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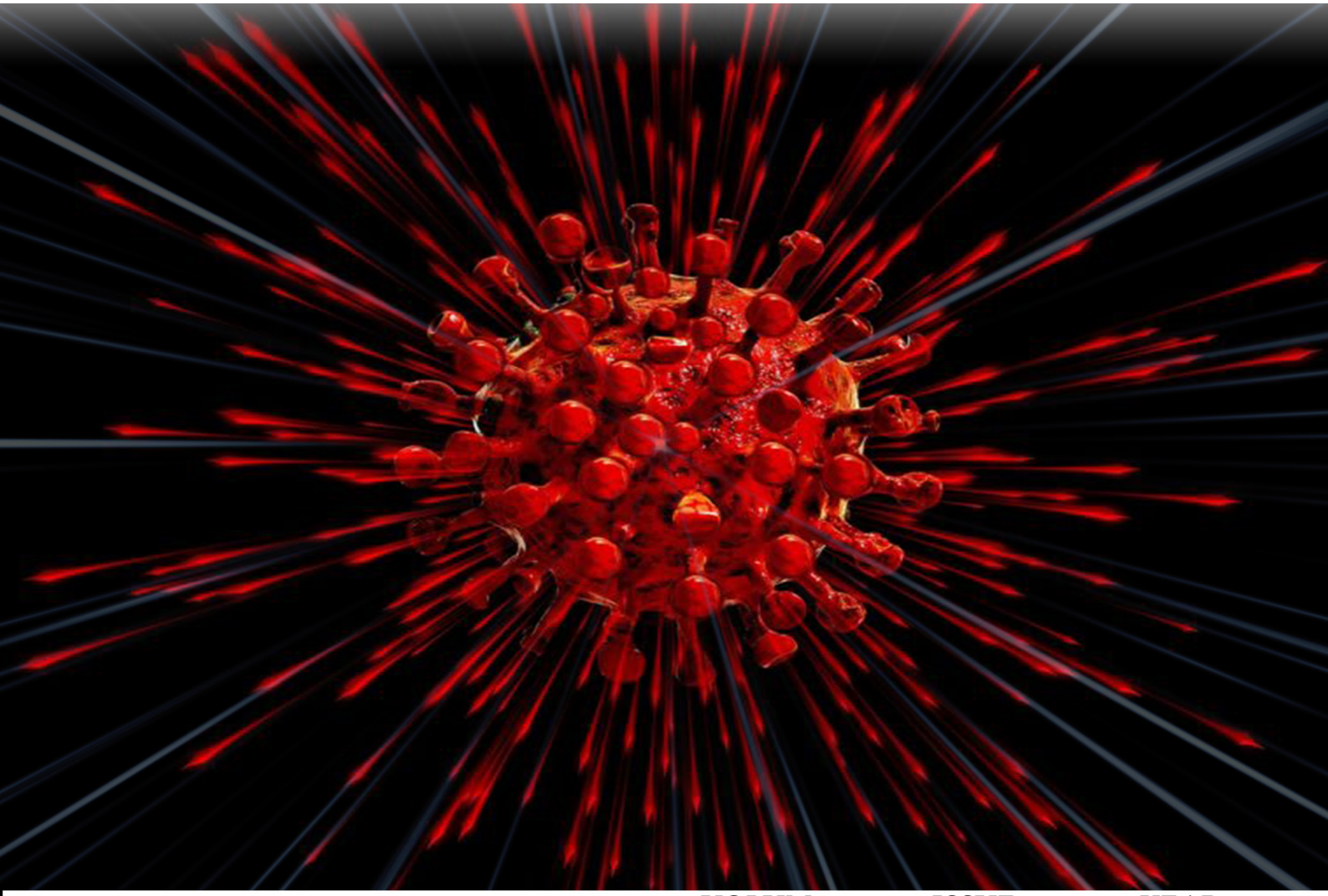

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The interest in our journal, and therefore both the number and quality of our articles, is increasing day by day. Recently, our journal **Anatolian Current Medical Journal** (**ACMJ**) entered strong indexes. In this issue, we have received 19 original articles from different fields of medicine, and an interesting case report that I believe will guide our colleagues. In addition, there are 6 interesting original articles on COVID-19 that are still up to date. After **Journal of Medicine and Palliative Care** (**JOMPAC)**, which recently entered TR-Dizin ULAKBİM, we believe that **ACMJ** will soon enter TR-Dizin ULAKBİM. We thanks our valuable colleagues who contributed as authors, and to everyone who contributed to the journal.

Best Regards

Prof. Aydın ÇİFCİ, MD
Editor-in-Chief

CONTENTS

Original Article

CONTENTS

Original Article

Case Report

ERRATUM

Original Article

ANATOLIAN CURRENT MEDICAL

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A retrospective look at influenza during the COVID-19 pandemic

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ABSTRACT

Aim: Influenza is the main cause of acute respiratory disease worldwide and is transmitted via the respiratory secretions of infected individuals. The aim of this study was to retrospectively investigate influenza, a virus with which we have a longer history, during a period in which the COVID-19 pandemic has dominated current events in terms of viral infections.

Material and Method: Epidemiological and laboratory data of patients over 18 years of age who tested positive for influenza infection and received inpatient treatment in the Erzurum Regional Training and Research Hospital between January 1, 2019 and December 31, 2020 according to the influenza management algorithm of the Public Health Institution of Turkey were evaluated retrospectively.

Results: Of the 164 patients included in our study, 129 were hospitalized due to influenza A and 35 due to influenza B. Procalcitonin, aspartate transaminase, alanine transaminase, creatine kinase, total bilirubin, and direct bilirubin levels were significantly higher in the influenza A group compared to the influenza B group (p=0.002, 0.005, 0.006, 0.030, 0.010, and 0.004, respectively). Ten of the patients in the study died; there was no significant difference in mortality based on influenza subtype or presence of comorbidity (p=0.999 and 0.756, respectively). Forty-one (54.7%) of the patients with comorbidities had received an influenza vaccine.

Conclusion: Although COVID-19 has dominated the global stage since the pandemic started, the effects of periodic pandemics of our old acquaintance influenza still continue. Vaccination, which is our strongest weapon against pandemics, can reduce mortality in patients with comorbidities, as seen in our study.

Keywords: Influenza, pandemic, comorbidities

INTRODUCTION

The 1918 "Spanish flu" pandemic killed more than 50 million people and is the deadliest epidemic in recorded human history. Subsequent influenza epidemics such as the Asian flu in 1957, Hong Kong flu in 1969, and swine flu in 2009 had lower mortality and morbidity. Because influenza virus is more stable at cold temperatures, outbreaks usually occur during the dry, cold winter months. Influenza viruses are enveloped RNA viruses from the Orthomyxoviridae family (1). Three different influenza viruses have been identified: influenza A, B, and C. Hemagglutinin and neuraminidase are their two main envelope glycoproteins. Due to the segmented genome structure of the genes encoding these glycoproteins, mutations and antigenic changes can lead to epidemics and pandemics (2). Influenza A shows the greatest antigenic shift and antigenic drift of the influenza viruses and is therefore the main driver of outbreaks (3).

Influenza is the leading cause of acute respiratory disease worldwide. It is transmitted via the respiratory secretions of infected people. Symptoms occur approximately 1 to 4 days after infection and patients are infectious from 1 day before to 5 to 7 days after symptom onset. The most common symptoms are chills, fever, sore throat, nasal congestion, generalized muscle aches, headache, and fatigue, with vomiting and diarrhea seen mostly in children. It can present with more severe clinical signs and symptoms in people with comorbidities, individuals over 65 years old, and pregnant female (4). In addition to the direct effect of the virus, the most

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common cause of morbidity and mortality in these patient groups is superinfections that cause pneumonia and acute respiratory distress. Therefore, vaccination is recommended for these groups before the influenza season (5).

Influenza diagnosis can be made by virus polymerase chain reaction (PCR) test or culture of respiratory secretions obtained by nose and throat swab within the first days of symptom onset. Serological testing can also be done to detect antibodies in the serum (6,7). Antiviral agents used for treatment can be effective when started within the first days of symptoms. These include the M2 protein inhibitors amantadine and rimantadine and the neuraminidase inhibitors zanamivir and oseltamivir (8,9).

The aim of this study was to retrospectively evaluate the epidemiological and clinical findings of influenza patients who presented to the infectious diseases outpatient clinic and emergency department and were treated on an inpatient basis.

MATERIAL AND METHOD

The study was carried out with the permission of Erzurum Training and Research Hospital, Noninvasive Clinical Ethics Committee (Decision No:2021/08-153) All procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki.

Study Design

Epidemiological and laboratory data of patients over 18 years of age who tested positive for influenza infection and received inpatient treatment in the Erzurum Regional Training and Research Hospital between January 1, 2019 and December 31, 2020 according to the influenza management algorithm of the Public Health Institution of Turkey were evaluated retrospectively. Influenza was diagnosed by real-time PCR testing of nasopharyngeal swab samples obtained from patients with consistent clinical findings.

Definitions and Treatment

Patients who presented to the emergency department or infectious diseases outpatient clinic with the following signs and symptoms were admitted for inpatient treatment: shortness of breath or difficulty breathing, change in vital signs (arterial hypotension, defined as systolic blood pressure <90 mmHg and diastolic blood pressure <60 mmHg or >40 mmHg decrease from previous blood pressure; respiratory rate above 30/min; heart rate >120/min; hypoxia, defined as oxygen saturation <92% on pulse oximetry), change in consciousness, and severe dehydration (peripheral pulse loss or attenuation, reduced skin turgor, inability to read blood pressure, and loss of more than 10% of body weight, characterized by sunken eyes). Droplet isolation precautions were implemented in the patient rooms.

During follow-up, the patients were evaluated according to intensive care admission criteria and those with indications continued treatment in the isolation intensive care unit. The following parameters were considered as criteria for admission to intensive care:

Major

1. Need for invasive mechanical ventilation
2. Septic shock requiring vasopressor therapy

Minor

1. Respiratory rate ≥30/min
2. PaO_2/FiO_2 ratio ≤250
3. Presence of multilobar infiltrates on chest x-ray
4. Confusion/disorientation
5. Uremia (blood urea nitrogen [BUN] ≥20 mg/dL)
6. Leukopenia (white blood cell count <4000/mm³)
7. Thrombocytopenia (platelet count <1 ×105/mm³)
8. Hypothermia (body temperature <36°C)
9. Hypotension requiring intensive fluid therapy

Patients with one major risk factor or three minor risk factors were treated in the isolation intensive care unit.

In addition to symptomatic treatment, inpatients in our clinic received oseltamivir, which was shown to be effective for influenza A and B, in accordance with the National Pandemic Plan guideline (10). As oseltamivir has substantial renal excretion, dose adjustments were made for patients with creatine clearance below 30 mL/min. Patients weighing more than 40 kg received oseltamivir 75 mg (capsule) twice daily for 5 days. Patients with glomerular filtration rate (GFR) of >30 to 60 mL/min received 30 mg (capsule or suspension) twice daily, and those with a GFR of >10 to 30 mL/min received a single daily dose of 30 mg (capsule or suspension). Patients undergoing hemodialysis or peritoneal received a single dose of 30 mg (capsule or suspension) after their dialysis session. The standard duration of treatment was 5 days, which was extended to 10 days for intensive care patients, immunocompromised patients, and those with severe infections. In patients whose procalcitonin level was above the upper limit (0.5 mg/dL) determined for our hospital, empirical antibiotic therapy for bacterial superinfection was started with the correlation of clinical findings. Antibiotic regimens were revised according to sputum, blood, and urine culture results.

Statistical Analysis

The data were analyzed using IBM SPSS Statistics for Windows version 20.0 (IBM Corp, Armonk, NY). Kolmogorov-Smirnov test were used to determine whether continuous variables were normally distributed. Pearson chi-square, Fisher's exact test and Mann–Whitney U test were used for comparisons of parametric data and non normally distributed numerical data, respectively, between groups. Independent-samples t test was used to compare demographic data and laboratory parameters between groups. Receiver operating characteristic (ROC) curve analysis was used to determine the diagnostic value of continuous variables and the Youden index was used to determine cut-off values. P-values lower than 0.05 were considered statistically significant.

RESULTS

The mean age of the 164 patients included in the study was 50.3±23.3 years. The mean ages of patients with influenza A and influenza B infection were 52.8±20.2 years and 41.1±18.4 years, respectively (p=0.002). Female accounted for 74 (57.4%) of the 129 patients with influenza A and 19 (54.3%) of the 35 patients with influenza B (p=0.744).

Seventy-five (45.7%) of the 164 patients in our study had comorbidities. The most common comorbidity was chronic obstructive pulmonary disease (COPD) (n=52, 69.3%), followed by coronary artery disease (n=32, 42.6%), hypertension (n=20, 26.7%), diabetes mellitus (n=13, 17.3%), and malignancy (n=12, 16%). The distribution of patients with and without comorbidities was 60 (46.5%) and 69 (53.5%) in the influenza A group and 15 (42.9%) and 20 (57.1%) in the influenza B group, respectively. There was no statistically significant difference in the frequency of comorbidity based on influenza type (p=0.700). In comparisons of age and laboratory data between patients with comorbidities compared to those without comorbidities, hemoglobin level was found to be significantly lower in patients with comorbidities (12.9±2.3 vs. 13.6±2.1, p=0.050). Other parameters showed no statistically significant differences between patients with and without comorbidities (p >0.05). The median value of hemoglobin level in surviving patients was 13.5, while it was 10 in non-survival patients. A statistically significant difference was observed between both groups (p= 0.001)

The laboratory data at time of admission in patients with influenza A and B are shown in **Table 1**. Procalcitonin, aspartate transaminase (AST), alanine transaminase (ALT), creatine kinase, total bilirubin, and direct bilirubin levels were significantly higher in influenza A group compared to influenza B group (p=0.002, 0.005, 0.006, 0.030, 0.010, and 0.004, respectively). In contrast, albumin level was significantly higher in the influenza B group (p=0.020).

The H1N1 subtype was detected in 96 (74.4%) of the patients with influenza A and the H3N2 subtype was detected in the other 33 patients (25.6%). Patients with H3N2 had a significantly higher mean age than those with H1N1 (58.2±20.9 years versus 50.4±19.7 years, p=0.050). No statistically significant differences in laboratory data were observed between the subtypes.

Ten patients died during follow-up, 8 (80%) in the influenza A group and 2 (20%) in the influenza B group. Mortality did not differ significantly between influenza A and B (p=0.999). The comparison of age and laboratory data of surviving and nonsurviving patients is shown in **Table 2**. Nonsurviving patients had significantly higher WBC count, neutrophil count, mean platelet volume, red cell distribution volume, CRP, erythrocyte sedimentation rate, procalcitonin, international normalized ratio, prothrombin time, activated partial prothrombin time, fibrinogen, D-dimer, ferritin, BUN, creatinine, AST, ALT, lactose dehydrogenase, creatine kinase, alkaline phosphatase, total bilirubin, direct bilirubin, neutrophil/lymphocyte ratio (NLR), and platelet/lymphocyte ratio (PLR) than surviving patients (p <0.050). Hemoglobin, lymphocyte count, total protein, and albumin levels were significantly lower in nonsurviving patients (p <0.001). Four of the nonsurviving patients (40%) had comorbidities and 6 (60%) did not. Statistical analysis revealed no significant association between comorbidity and mortality (p=0.756).

In the ROC curve analysis between surviving and nonsurviving patients, the areas under the curve for procalcitonin, CRP, fibrinogen, D-dimer, and ferritin were 0.886, 0.847, 0.784, 0.925, and 0.763, respectively. When these variables were evaluated based on Youden index cut-off values, sensitivity and specificity respectively were 90% and 79% for procalcitonin at a cut-off of 1.3 ng/mL; 80% and 77% for CRP at a cut-off of 53.1 mg/L; 80% and 63% for fibrinogen at a cut-off of 413.5 ng/mL; 90% and 88% for D-Dimer at a cut-off of 1630 ng/mL; and 90% and 60% for ferritin at a cut-off of 155.5 ng/ml (**Figure 1**).

Due to the global problem in influenza vaccine shipments due to the COVID-19 pandemic, only people over 65 years of age and patients with comorbidities were vaccinated in our country during the study period. The vaccination rate among the 75 patients with comorbidities in our study was 54.7% (n=41). Two of the 41 vaccinated patients died.

Table 1. Comparison of age and laboratory parameters between patients with influenza A and B

	Influenza A (n=129) Median (IQR) (min.-max)	Influenza B (n=35) Median (IQR) (min.-max)	p
Age (years)	62 (38.1-81.2) (18-94)	42 (26.4-69.5) (19-92)	0.002
WBC (/µL)	6450 (3156-14580) (2050-27780)	7300 (2630-14230) (2590-18760)	0.710
Hemoglobin (g/dl)	12.8 (9.7-15.4) (8-17)	13.6 (10.1-16.9) (7.6-18.5)	0.770
Neutrophils (/µL)	4870 (810.4-10100.4) (790-25830)	5390 (850.4-91601) (1520-14210)	0.830
Lymphocytes (/µL)	880 (954-1640) (100-3160)	1240 (765.9-1890.6) (360-3570)	0.070
Monocytes (/µL)	440 (50-890) (10-1200)	600 (98-980) (100-4680)	0.118
Eosinophils (/µL)	10 (0-140) (0-550)	10 (0-135) (0-330)	0.633
Basophils (/µL)	10 (0-30) (0-70)	20 (0-45) (0-90)	0.460
Platelets (/µL)	187000 (154000-264000) (40000-620000)	223000 (210000-286000) (27000-368000)	0.670
MPV (fL)	10.6 (8.9-12.1) (8.6-13.9)	9.9 (9.1-11.7) (9.2-12.4)	0.280
RDW	13.8 (11.7-16.4) (11.9-29.3)	13.1 (10.1-16.4) (11.6-21.2)	0.610
CRP (mg/L)	36.7 (16.9-92.4) (3-396)	13.3 (18.4-88.9)(3-231)	0.790
Sedimentation (s)	28 (5-43) (2-96)	17 (2-37) (2-90)	0.240
Procalcitonin (ng/mL)	0.63 (0.45-5.65) (0.01-32.79)	0.03 (0.06-1.14) (0-3.5)	0.002
INR	1.12 (0.95-1.14) (0.9-1.9)	1.13 (0.99-1.2) (0.8-2.0)	0.560
PT (s)	14.6 (12.1-16.7)(11.3-33)	14.2 (12.6-15.8) (13.2-19.9)	0.560
aPTT (s)	29.4 (20.8-26.4) (21.2-74.9)	31 (21.1-26.9) (21.9-43.8)	0.940
Fibrinogen (ng/mL)	410 (210-580.4) (174-735)	352 (225-590) (131-918)	0.780
D-dimer (ng/mL)	1000 (450-2560) (100-14000)	420 (340-2460) (147-4600)	0.110
Ferritin (ng/ml)	156 (80.4-456.9) (10.2-1600)	124 (76.4-637.4) (15-1289)	0.520
BUN (mg/dL)	20.1 (8.6-35.5) (4.7-104)	15 (7.9-36.1) (9.9-119)	0.200
Creatine (mg/dL)	0.96 (0.6-1.8) (0.54-8.9)	0.81 (0.55-1.89) (0.19-4.99)	0.160
AST (U/L)	35 (33.4-101.6) (11-1735)	22 (19-41.6) (14-53)	0.005
ALT (U/L)	30 (10-78.4) (6-1164)	20 (12-36.6) (6-53)	0.006
LDH (U/L)	234 (167.8-510.8) (165-2929)	303 (142.4-470.4) (183-638)	0.230
CK (U/L)	128 (45.4-468.9) (22-13649)	80 (34.4-130.4) (29-243)	0.030
GGT (U/L)	32 (19-120.9) (11-959)	21 (14-110.6) (10-121)	0.140
ALP (U/L)	77 (42-94.6) (33-633)	82 (37-95.5) (45-165)	0.370
Total bilirubin (mg/dL)	0.5 (0.3-1.2) (0.1-3.8)	0.4 (0.3-0.8) (0.25-1.72)	0.010
Direct bilirubin (mg/dL)	0.23 (0.15-0.99) (0.1-1.17)	0.17 (0.15-0.35) (0.1-0.54)	0.004
Total protein (g/dl)	6.1 (4.7-7.2) (4.5-9.3)	7.2 (4.6-7.1) (4.8-8.1)	0.790
Albumin (g/dl)	3.4 (2.34-3.6)(2.21-4.6)	4.1 (3.6-4.6) (1.8-4.8)	0.020
NLR	5.18 (3.48-7.65) (0.47-88.71)	3.42 (2.98-6,96) (0.7-34.5)	0.330
PLR	170.3 (56.8-190.6) (44.9-3100)	191.2 (55.4-110.6) (7.56-603.3)	0.280

WBC: White blood cells, MPV: Mean platelet volume, RDW: Red blood cell distribution width, INR: International normalized ratio, PT: Prothrombin time, aPTT: Activated partial prothrombin time, NLR: Neutrophil/lymphocyte ratio, PLR: Platelet/lymphocyte ratio, AST: Aspartate aminotransferase, ALT: Alanine aminotransferase, LDH: Lactate dehydrogenase, GGT: Gamma glutamyl transferase, ALP: Alkaline phosphatase, BUN: Blood urea nitrogen, CK: Creatine kinase, SD: Standard deviation, p: Comparison of parameters between groups

DISCUSSION

Our retrospective analysis of influenza patients during the COVID-19 pandemic showed that 129 of 164 patients had influenza A and 35 had influenza B infections. The mean age was higher among influenza A patients than influenza B patients. Liver function tests and procalcitonin levels were also higher among influenza A patients. Influenza A subtype analysis revealed a higher rate of H1N1 compared to H3N2, while H3N2 was more frequent in older patients. In the analysis of the 10 patients who died, the difference in mortality between patients with influenza A and B was not statistically significant. However, procalcitonin, CRP, fibrinogen, and D-dimer levels were significantly higher in nonsurviving patients than surviving patients.

The annual recurrent respiratory disease caused by influenza viruses has a significant impact on human health and the economy. Although influenza B viruses (IBV) are found almost exclusively in humans, influenza A viruses (IAVs) circulate in annual outbreaks within the human population and emerge from a large zoonotic reservoir (11). In a process called antigenic drift, IAVs can rapidly acquire adaptive mutations, enabling them evade the immune system response. In addition, their multiple RNA segments facilitate the reassortment of genetic elements from different IAVs. This process, called antigenic shift, is responsible for the increased pathogenicity and infectivity of the virus in pandemic outbreaks. In addition, studies have shown that the interaction of hemagglutinin (HA) and neuraminidase (NA) has an important place in the replication of the

Table 2. Comparison of age and laboratory parameters between surviving and nonsurviving influenza patients

	Survivors (n=154) Median (IQR) (min.-max)	Non-survivors (n=10) Median (IQR) (min.-max)	p
Age (years)	53 (22-48) (18-94)	70 (64-78) (21-92)	0.002
WBC (/µL)	6475 (4530-8210) (2050-27780)	9970 (8450-12690) (1650-19800)	0.030
Hemoglobin (g/dl)	13.5 (12.4-14.6) (8-18.5)	10 (8.8-12.1) (7.6-13.7)	0.001
Neutrophils (/µL)	4585 (2880-7140) (790-25830)	8305 (6500-9670) (2520-18090)	0.005
Lymphocytes (/µL)	985 (756-1103) (250-3570)	765 (540-910) (100-1430)	0.001
Monocytes (/µL)	515 (310-796) (70-4680)	450 (356-750) (10-870)	0.390
Eosinophils (/µL)	10 (5-90) (0-550)	0 (0-100) (0-260)	0.660
Basophils (/µL)	20 (10-40) (0-90)	15 (0-30)(0-40)	0.450
Platelets (/µL)	205000 (196000-260000) (27000-456000)	207500 (180500-250000) (9100-620000)	0.890
MPV (fL)	10.1 (8.9-11) (8.6-13.9)	11 (10.5-11.8) (10.2-12.7)	0.020
RDW	13.3 (12.1-18.6) (11.6-29.3)	15.6 (14.3-16.5) (13.2-17.1)	0.020
CRP (mg/L)	17 (10-33) (3-396)	93 (80-135) (31.1-290)	0.001
Sedimentation (s)	21 (15-33) (2-96)	48.5 (42-66) (7-90)	0.001
Procalcitonin (ng/mL)	0.1 (0-0.6) (0-12.3)	5.5 (0.9-2.4) (0.1-32.8)	0.001
INR	0.9 (0.8-1) (0.8-1.5)	1.9 (1.5-2) (1.4-2)	0.020
PT (s)	14.3 (13.5-16.4) (11.3-33)	17.7 (16.8-20.4) (13-21.9)	0.020
aPTT (s)	29.4 (28.4-35.4) (21.9-74.9)	32.5 (30.4-39.6) (21.2-73.3)	0.020
Fibrinogen (ng/mL)	373 (188-299) (131-918)	503 (430-667) (341-728)	0.006
D-dimer (ng/mL)	636.5 (225-860) (100-14000)	3185 (2456-6530) (1260-8490)	0.005
Ferritin (ng/ml)	129.3 (88-260) (10.2-1289)	249 (210-480) (130-1600)	0.001
BUN (mg/dL)	15.7 (12.6-36.4) (4.7-104)	43.3 (29.6-49.8) (9-119)	0.010
Creatine (mg/dL)	0.9 (0.8-1.4) (0.2-8.9)	1.9 (1.7-3.2) (0.6-4.99)	0.020
AST (U/L)	28 (22-45) (13-681)	49 (41-102) (11-1735)	0.001
ALT (U/L)	26.5 (20-47) (6-292)	62 (55-136) (6-1164)	0.001
LDH (U/L)	299.5 (210-350)(165-2156)	444 (360-596) (196-2929)	0.001
CK (U/L)	97.5 (80-106.5) (22-2359)	173.5 (150.6-360.6) (72-13649)	0.001
GGT (U/L)	28 (20-45) (10-242)	72 (50-96) (16-959)	0.001
ALP (U/L)	74.5 (65-95) (40-192)	150.5 (110-230) (33-633)	0.001
Total bilirubin (mg/dL)	0.5 (0.4-0.9) (0.1-3.04)	0.6 (0.5-2.2) (0.2-3.8)	0.007
Direct bilirubin (mg/dL)	0.21 (0.1-0.4) (0.1-1.17)	0.25 (0.2-0.9) (0.1-1.1)	0.040
Total protein (g/dl)	6.5 (5.1-6.8) (4.5-9.3)	5.6 (4.8-5.9) (4.6-7.7)	0.001
Albumin (g/dl)	3.7 (3.4-4.1) (2.3-4.8)	3.1 (2.7-3.3) (1.8-4)	0.001
NLR	3.6 (2.8-5.6) (0.5-34.5)	15.3 (10.2-19.8) (3.5-88.7)	0.001
PLR	171.2 (160-200.4) (7.6-854.6)	296.6 (256-410) (80.4-3100)	0.001

WBC: White blood cells, MPV: Mean platelet volume, RDW: Red blood cell distribution width, INR: International normalized ratio, PT: Prothrombin time, aPTT: Activated partial prothrombin time, NLR: Neutrophil/lymphocyte ratio, PLR: Platelet/lymphocyte ratio, AST: Aspartate aminotransferase, ALT: Alanine aminotransferase, LDH: Lactate dehydrogenase, GGT: Gamma glutamyl transferase, ALP: Alkaline phosphatase, BUN: Blood urea nitrogen, CK: Creatine kinase, SD: Standard deviation, p: Comparison of parameters between groups

virus. It has been observed that the virus mutates in order to continue its replication in host cells where balanced HA and NA activities are not realized. While these mutations facilitate the adaptation of the virus to the hosts, it also enabled it to escape from antiviral treatments. The antigenic properties of HA and NA on IAVs enable them to be divided into subtypes, the most common of which are H1N1 and H3N2 (12,13).

Influenza virus infections primarily cause uncomplicated respiratory tract infection characterized by fever, muscle aches, chills and shivering, and fatigue lasting approximately 2 to 8 days. Onset is rapid, and it may also present with gastrointestinal symptoms such as vomiting and diarrhea in the pediatric age group (14). Older patients, pediatric patients, and patients with high comorbidity can develop viral or secondary bacterial pneumonia that causes respiratory and multiple organ failure (15). Therefore, in addition to patients receiving immunosuppressive therapy, patients with obstructive pulmonary disease, congenital or acquired heart disease, chronic kidney and liver disease, and individuals over 65 years of age are risk groups for clinically severe illness (16). This increases the importance of inducing active immunity with vaccination in these populations before outbreaks occur (17-20).

Most of the patients in our study had IAV infection, and patients with IAV infection were statistically older than those with IBV infection. In addition, patients with IAV infection had higher procalcitonin levels and liver function tests at admission. These results suggest that IAV, which mutates more frequently than IBV, is

more easily transmitted to all age groups and may cause more severe illness in elderly patients in particular due to both its direct effect and the superinfections it can lead to. In the IAV subtype analysis, we observed that H1N1 was more common than H3N2.

Of the 10 patients who died, 8 had IAV infection and 2 had IBV infection. The nonsurviving patients showed significantly greater changes in nearly all studied laboratory parameters when compared with surviving patients. This may also be evidence that IAV infection leads to more severe clinical manifestations due to its high frequency among older patients and the associated superinfections that occur in this patient population. In this study, there was no statistically significant difference in mortality based on the presence of comorbidities. This result may be related to the fact that 41 of the 75 patients with comorbidities in our study with influenza vaccine priority during the pandemic (due to age and presence of comorbidities) were vaccinated. We observed with influenza the most concrete evidence of what we have experienced so bitterly during the current COVID-19 pandemic, that severe clinical illness can be prevented with vaccination. In addition, in the ROC curve analysis of nonsurviving patients, the high areas under the curve and sensitivity of procalcitonin, CRP, fibrinogen, D-dimer, and ferritin may indicate that these parameters can be used to predict mortality, as in the COVID-19 pandemic.

The main limitation of our study was the inability to observe the effect of comorbidities on mortality and laboratory parameters due to the lack of homogeneity in the vaccinated patient population. The priority given to the at-risk population in the vaccination program during the pandemic is a factor that prevented an increase in mortality in these patients. Furthermore, as superinfections cannot be predicted in advance and patients are given empiric antibiotherapy based on their laboratory results, it is not clear whether changes in laboratory parameters occurred due to influenza or superinfections that developed afterwards.

In conclusion, IAV infection occurs more frequently than IBV and may present with more severe disruptions in laboratory parameters. The IAV subtype H1N1 was found to cause infection more frequently in our region. The high procalcitonin levels observed in nonsurviving patients suggest superinfections as the likely precipitating factor of mortality in these patients. Our analysis of the relationship between comorbidity and mortality indicated that the vaccine policy prioritizing adults over 65 years of age and those with comorbidities had a favorable impact on mortality.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Erzurum Training and Research Hospital, Noninvasive Clinical Ethics Committee (Decision No:2021/08-153)

Informed Consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

Financial Disclosure: The authors declared that this study has received no financial support.

Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper and that they have approved the final version

REFERENCES

1. Hutchinson EC. Influenza virus. Trends Microbiol 2018; 26: 809-10.
2. Palese P, Young JF. Variation of influenza A, B, and C viruses. Science 1982 ; 215: 1468-74.
3. Webster RG. Antigenic variation in influenza viruses. Origin and evolution of viruses: Elsevier; 1999. p. 377-90.
4. Madjid M, Miller CC, Zarubaev VV, et al. Influenza epidemics and acute respiratory disease activity are associated with a surge in autopsy-confirmed coronary heart disease death: results from 8 years of autopsies in 34 892 subjects. Eur Heart J 2007; 28: 1205-10.
5. Jackson ML, Chung JR, Jackson LA, et al. Influenza vaccine effectiveness in the United States during the 2015–2016 season. New Engl J Med 2017; 377: 534-43.
6. Ellis JS, Zambon MC. Molecular diagnosis of influenza. Rev Med Virol 2002; 12: 375-89.
7. Wiselka M. Influenza: diagnosis, management, and prophylaxis. BMJ 1994; 308: 1341-5.
8. Hayden FG, de Jong MD. Emerging influenza antiviral resistance threats. Oxford University Press; 2011.
9. Oh DY, Hurt AC. Using the ferret as an animal model for investigating influenza antiviral effectiveness. Front Microbiol 2016; 7: 80.
10. Republic of Turkey MoH. National Action Plan in Pandemic Influenzae. Ankara2006. p. 77-82.
11. Mostafa A, Abdelwhab EM, Mettenleiter TC, Pleschka S. Zoonotic potential of influenza A viruses: a comprehensive overview. Viruses 2018; 10: 497.
12. Gamblin SJ, Skehel JJ. Influenza hemagglutinin and neuraminidase membrane glycoproteins. J Biol Chem 2010; 285: 28403-9.
13. Mitnaul LJ, Matrosovich MN, Castrucci MR, et al. Balanced hemagglutinin and neuraminidase activities are critical for efficient replication of influenza A virus. J Virol 2000; 74: 6015-20.
14. Monto AS, Gravenstein S, Elliott M, Colopy M, Schweinle J. Clinical signs and symptoms predicting influenza infection. Arch Intern Med 2000; 160: 3243-7.

15. Van Reeth K, Brown IH, Dürrwald R, et al. Seroprevalence of H1N1, H3N2 and H1N2 influenza viruses in pigs in seven European countries in 2002–2003. Influenza and other respiratory viruses 2008; 2: 99-105.
16. Kerget B, Çelik E, Kerget F, Aksakal A, Uçar EY, Araz Ö, et al. Evaluation of 3-month follow-up of patients with post-acute COVID-19 syndrome. J Med Virol 2022; 94: 2026-34.
17. Wilhelm M. Influenza in older patients: a call to action and recent updates for vaccinations. Am J Manag Care 2018; 24: S15-S24.
18. Zimmerman RK, Santibanez TA, Janosky JE, et al. What affects influenza vaccination rates among older patients? An analysis from inner-city, suburban, rural, and Veterans Affairs practices. Am J Med 2003; 114: 31-8.
19. Güngörer B. Baseline demographic, clinical and laboratory risk factors for predicting admission to intensive care unit in patients diagnosed with COVID-19 in the emergency department. Anatolian Curr Med J 2021; 3: 279-83.
20. Kerget B, Kerget F, Aksakal A, Aşkın S, Uçar EY, Sağlam L. Evaluation of the relationship between KIM-1 and suPAR levels and clinical severity in COVID-19 patients: A different perspective on suPAR. J Med Virol 2021; 93: 5568-73.

Original Article

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Ramadan and health: a scientometric analysis of health literature on Ramadan and fasting

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ABSTRACT

Aim: During Ramadan month, every year, approximately two billion of Muslims practice fasting and avoid eating, drinking and intercourse from dawn to dusk throughout the world. Although the effects of Ramadan fasting on human health were highly studied in academic literature, there are only limited number of scientometric articles referring to Ramadan and health.

Material and Method: We performed a scientometric analysis of "Ramadan and health" publications indexed in Web of Science databases between 1980 and 2019 and found a total of 497 articles.

Results: The most published documents were original articles (88.13%). The most studies areas were found to be Religion, Nutrition and Endocrinology, (n=269, 214 and 184 items, respectively). The USA was leading country with 76 items followed by Saudi Arabia, the UK, Turkey and Iran (n=76, 58, 55, 39 and 36 papers, respectively). King Saud University (Saudi Arabia) ranked first in institutions with 21 items. H-index of Ramadan and health literature was measured as 40 and total number of citations was 5837. The most indexed keywords were "Ramadan", "fasting", "diabetes", and "pregnancy". The USA, the UK, Saudi Arabia and Canada were found as the most collaborative countries.

Conclusion: The importance of scientometric studies has been increasing in recent years. We think that this scientometric study data about Ramadan and fasting which are the conditions of the religion of Islam will contribute to scientists.

Keywords: Ramadan, health, fasting, scientometrics, bibliometrics

INTRODUCTION

Diurnal fasting for a limited number of days is ordained in Islam and Judaism in Abrahamic religions. In Islam, fasting during the month of Ramadan was fundamentally ordered for Muslims according to the surah al-Baqarah verse 183-184 in Holy Quran (1). During Ramadan month, every year, nearly two billion of Muslims practice Ramadan fasting and abstain from eating, drinking and intercourse from dawn to sunset throughout the world (2). Scientometrics is a statistical discipline evaluating academic literature in a certain field (3). Although the effects of Ramadan fasting on human health were highly investigated in academic literature, there are only limited number of scientometric articles referring to Ramadan and health.

MATERIAL AND METHOD

Ethics committee approval was not required for the preparation of the article. Institutional approval was obtained for the preparation of the article.

We analyzed all articles in the literature of Ramadan and health indexed in Web of Science (WoS, Clarivate analytics, USA) databases titled Core Collection, SciELO Citation Index, Russian Science Citation Index and Korean Journal Database. All papers published between 1980 and 2019 were included in our analysis. A search string containing keywords of "Ramadan" and "health" were used for our scientometric search. A free web source named GunnMap 2 was used to generate global productivity map in academic literature (4).

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Scientometric network images were created with a free software tool titled VOSviewer (version 1.6.7, Copyright Centre for Science and Technology Studies, Leiden University, The Netherlands) (5).

RESULTS

General features

Our search yielded 497 articles and only 35.61% of which were open access. Main language of the literature was English (96.98%) followed by French, Arabic, Spanish and German (3.22, 1.61, 1.61 and 0.4%, respectively). Original articles covered 88.13% of all literature (**Table 1**). The most studies areas were found to be Religion, Nutrition, Endocrinology, Psychology and Health Care (n=269, 214, 184, 178 and 150 documents, respectively; **Table 2**). The peak year of the literature was 2019 with 73 items. 1982 was the first year that articles published in this area (**Figure 1**). The first original article in Ramadan and health literature was titled "Changes in certain blood-constituents during Ramadan" written by Fedail SS et al. (6) and published in American Journal of Clinical Nutrition.

Table 1. Types of documents published on health literature pertaining to Ramadan and health*

Document type	Number	%a
Original article	438	88.129
Review	68	13.682
Meeting report	47	9.457
Abstract	35	7.042
Editorial material	20	4.024
Letter	14	2.817
Clinical trial	11	2.213
Other/Unspecified	9	1.811
Case report	4	0.805
Book	2	0.402
Biography	1	0.201
Total	497	100

Table 2. The top ten research areas of documents in Ramadan and health literature according to Web of Science database between 1980 and 2019

Research Areas	Number of publications	%
Religion	269	54.125
Nutrition/Dietetics	214	43.058
Endocrinology	184	37.022
Psychology	178	35.815
Health Care	150	30.181
Environmental/Occupational Health	145	29.175
Behavioral Sciences	141	28.370
Pathology	127	25.553
Internal Medicine	119	23.944
Pharmacology	116	23.340

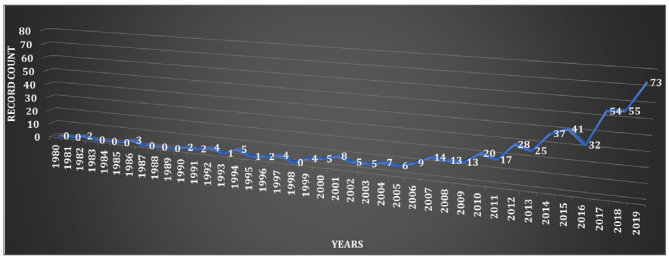

Figure 1. Record count of Ramadan and health literature by year

Countries, authors, institutions and source titles

The most contributor countries were the United States of America (USA), Saudi Arabia, the United Kingdom (UK), Turkey and Iran (n=76, 58, 55, 39 and 36 documents, respectively; **Figure 2**). Publications were distributed to 61 countries (**Figure 3**). The most prolific author was Hassanein M with 10 articles from University of Leicester (UK) (**Table 3**). King Saud University (Saudi Arabia) was the leading institution with 21 items (**Table 4**). The most productive journal in this field were Diabetes Research and Clinical Practice, Journal of The Pakistan Medical Association and Journal of Sports Sciences (4.22, 2.21 and 1.81%, respectively; **Table 5**). According to WoS Core Collection, top ten funding agencies were German Research Foundation (Germany), National Institutes of Health (USA) and The United States Department of Health & Human Services (USA).

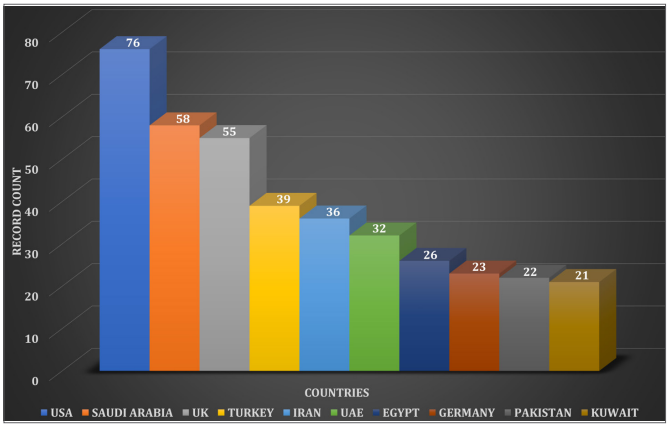

Figure 2. Top ten countries producing articles in Ramadan and health field by total record count

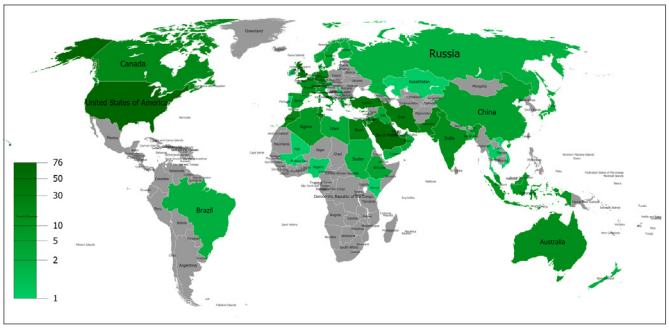

Figure 3. Global productivity in Ramadan and health literature

Table 3. The most prolific authors by record count in Ramadan and health literature

Author	Institution	Country	Record Count	%*
Hassanein M	University of Leicester	UK	10	2.012
Dvorak J	Schulthess Klinik	Switzerland	7	1.408
Van Ewijk R	Gutenberg University	Germany	7	1.408
Beshyah SA	Dubai Medical College	UAE	6	1.207
Bragazzi NL	Genoa University	Italy	6	1.207
Chamari K	AHP Research Center	Qatar	6	1.207
Maughan RJ	Loughborough University	UK	6	1.207
Wilbur K	Qatar University	Qatar	6	1.207
Zerguini Y	FIFA Medical Center of Excellence	Algeria	6	1.207
Leiper JB	Loughborough University	UK	5	1.004

Table 4. The top ten institutions by number of publications in literature of Ramadan and health literature

Organizations	Country	Document number	%
King Saud University	Saudi Arabia	21	4.217
Aspetar Orthopaedic Sports Medicine Hospital	Qatar	11	2.209
Dubai Hospital	UAE	11	2.209
King Abdulaziz University	Saudi Arabia	11	2.209
United Arab Emirates University	UAE	11	2.209
Harvard University	USA	10	2.008
Johannes Gutenberg University of Mainz	Germany	10	2.008
Kuwait University	Kuwait	10	2.008
University of Hassan II Casablanca	Morocco	8	1.606
Sackler Faculty of Medicine	Israel	8	1.606

Table 5. The first ten source titles according to the number of published documents in the literature of Ramadan and health

Journal Name	Number of Publications	%
Diabetes Research and Clinical Practice	21	4.217
Journal of The Pakistan Medical Association	11	2.209
Journal of Sports Sciences	9	1.807
Saudi Medical Journal	8	1.606
International Journal of Clinical Pharmacy	7	1.406
American Journal of Epidemiology	6	1.205
Annals of Nutrition and Metabolism	6	1.205
Diabetes	6	1.205
Eastern Mediterranean Health Journal	6	1.205
Nutrients	6	1.205

Citation report

H-index of Ramadan and health literature was measured as 40 and total number of citations was 5837 (4356 without self-citations). Average citations per item were calculated as 11.72. The peak year was 2019 with 929 citations. The most cited manuscript was an original article investigating relation between diurnal fasting in Ramadan and fetal health, titled "Health Capital and the Prenatal Environment: The Effect of Ramadan Observance During Pregnancy" published in 2011 (7) (**Table 6**).

Table 6. The ten most cited manuscripts in the literature of Ramadan and health

Article	Author	Journal name/published	Total citation	Average citations per year
Health capital and the prenatal environment: the effect of ramadan observance during pregnancy	Almond, Douglas; Mazumder, Bhashkar	American Economic Journal-Applied Economics	154	15.40
Effects on health of fluid restriction during fasting in Ramadan	Leiper, JB; Molla, AM; Molla, AM	European Journal of Clinical Nutrition	127	7.06
A controlled trial of reduced meal frequency without caloric restriction in healthy, normal-weight, middle-aged adults	Stote, Kim S.; Baer, David J.; Spears, Karen; et al.	American Journal of Clinical Nutrition	124	8.86
The impact of religious fasting on human health	Trepanowski, John F.; Bloomer, Richard J.	Nutrition Journal	118	10.73
Ramadan Education and Awareness in Diabetes (READ) programme for Muslims with Type 2 diabetes who fast during Ramadan	Bravis, V.; Hui, E.; Salih, S.; et al.	Diabetic Medicine	113	10.27
Fenton's peroxidation and coagulation processes for the treatment of combined industrial and domestic wastewater	Badawy, M. I.; Ali, M. E. M.	Journal of Hazardous Materials	111	7.40
Physiological and chronobiological changes during Ramadan intermittent fasting	Roky, R; Houti, I; Moussamih, S; et al.	Annals of Nutrition and Metabolism	108	6.35
Impact of caloric and dietary restriction regimens on markers of health and longevity in humans and animals: a summary of available findings	Trepanowski, John F.; Canale, Robert E.; Marshall, Kate E.; et al.	Nutrition Journal	103	10.30
Daytime alertness, mood, psychomotor performances, and oral temperature during Ramadan intermittent fasting	Roky, R; Iraki, L; HajKhlifa, R; et al.	Annals of Nutrition and Metabolism	93	4.43
Diabetes and Ramadan: review of the literature	Benaji, B.; Mounib, N.; Roky, R.; et al.	Diabetes Research and Clinical Practice	89	5.93

Scientometric Network Analysis

The most indexed keywords were noted to be "Ramadan", "fasting", "diabetes", "pregnancy" and "Islam" (**Table 7**). Scientometric network map of the keywords revealed a flower pattern in which the keywords of "Ramadan" and "fasting" centered (**Figure 4**). The USA, the UK, Saudi Arabia, Canada and Germany were found to be the most cooperative countries (**Figure 5**). The most collaborative institutions in Ramadan and health literature were noted to be King Abdulaziz University (Saudi Arabia), University of Amsterdam (Netherlands), University of Genoa (Italy), Case Western Reserve University (USA) and Johannes Gutenberg University Mainz (Germany).

Table 7. Most used 20 keywords on the literature related to Ramadan and health literature

Keyword (Total link strength)	
1. Ramadan (389)	11. Pharmacist (30)
2. Fasting (323)	12. Saudi Arabia (27)
3. Diabetes (101)	13. Sleep (27)
4. Pregnancy (77)	14. Caloric restriction (25)
5. Islam (64)	15. Adherence (24)
6. Type 2 diabetes (58)	16. Insulin (23)
7. Hypoglycemia (44)	17. Intermittent fasting (23)
8. Nutrition (38)	18. Blood pressure (22)
9. Muslim (37)	19. Diet (22)
10. Quality of life (31)	20. Oxidative stress (21)

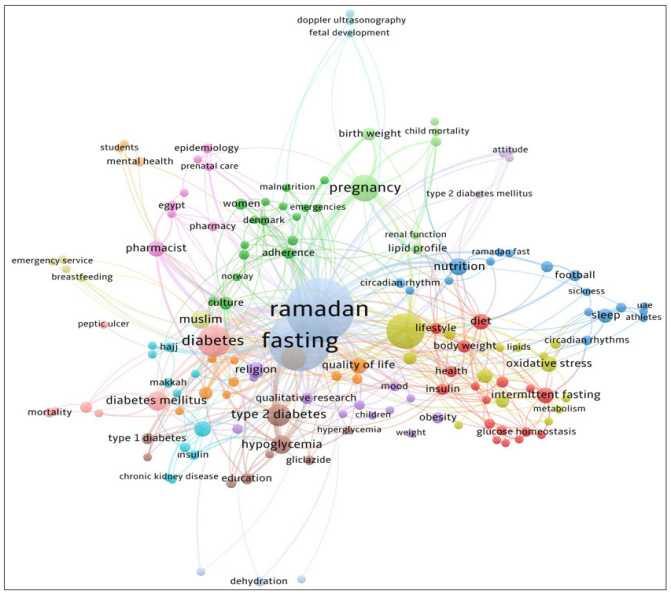

Figure 4. Scientometric network map of the most used keywords in Ramadan and health literature

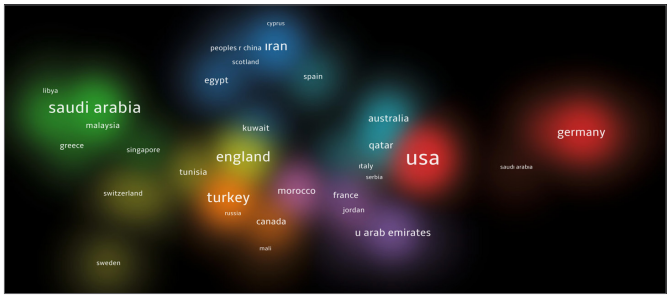

Figure 5. Scientometric map of the most collaborative countries in Ramadan and health literature

DISCUSSION

Scientometrics also known as "science of science" is an in-depth assessment of various quantitative and qualitative aspects of scientific literature in a certain field. Although the first scientometric study in the literature was conducted by Campbell in 1896, popularity of scientometrics has been increased in the last decades (8). In 1969, Pritchard proposed the term of "bibliometrics" instead of "statistical bibliography" and since then both terms of scientometrics and bibliometrics have been used interchangeably (9).

Beshyah and Beshyah performed a bibliometric analysis of the literature on Ramadan fasting and diabetes by searching Scopus database for a period of 1989 to 2018. The found a total of 424 and 112 items were open access. Original articles were 67% of all documents and reviews were 15.1%. The percentage of original articles was noted higher in our study (88.13%). The authors found that the UK ranked first in the literature followed by Saudi Arabia although the USA stood out in our analysis. Imperial College London and associated institutions jointly with Hamad Medical Corporation of Qatar were the most contributor institutions related to Ramadan and diabetes literature (10).

Husain et al. performed a bibliometric analysis of top cited 100 articles on literature of Ramadan and health indexed in WoS databases. The UK ranked first in publication of the most cited 100 articles with 15 items followed by Turkey, Tunisia, Saudi Arabia and Iran (n=13, 12, 11 and 6 documents,

respectively). Interestingly, this study had no data of the most cited articles and therefore we could not extract any document title to compare the results of our analysis (11).

Nouira et al. reported a bibliometric study investigating Ramadan and health literature indexed in Medline (PubMed) database. A total of 508 documents from 38 countries were noted. English was main language and 94% of all items were original articles. Saudi Medical Journal which was the fourth most contributor journal in our analysis ranked first with 19 items in the Medline indexed literature of Ramadan and health. The UK had the most indexed publications with 146 documents although we found the USA was leading country in our analysis.

Our study has one limitation that we analyzed all documents in only one database. Since WoS is the world's most trusted publisher-dependent database including global publications and citations we decided to perform or scientometric study by using WoS databases (12,13).

CONCLUSION

As a result, we think that other scientometric studies should be done in which articles written for Ramadan and some specific chronic diseases are evaluated.

ETHICAL DECLARATION

Ethics Committee Approval: Ethics committee approval was not required for the preparation of the article. Institutional approval was obtained for the preparation of the article.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: All authors declare that they have no conflict of interest.

Financial Disclosure: There is no person/organization that supports the study financially and the authors do not have any interest-based relationship

Author Contributions: Eskin F: Project, development, manuscript reviewing, data collection, editing and revising Şenel E: Project, development, data collection and analysis, writing, editing and revising

REFERENCES

1. Surah Al-Baqarah [2:183-193] - Al-Qur'an al-Kareem - القرآن الكريم [Internet]. [cited 2020 May 30]. Available from: https://quran.com/2/183-93
2. Khan MH, Al-Lehebi R. Respiratory disease and Ramadan. Lancet Respir Med 2020; 8: 449–50.
3. Şenel E. Health and ancient beliefs: A scientometric analysis of health literature related to Shamanism, Paganism and Spirituality. J Relig Health 2019; 58: 2019–35.
4. GunnMap. GunnMap 2 [Internet]. [cited 2020 Jan 10]. Available from: http://lert.co.nz/map/
5. Vosviewer. VOSviewer - Visualizing scientific landscapes [Internet]. [cited 2020 Jan 10]. Available from: https://www.vosviewer.com/
6. Fedail SS, Murphy D, Salih SY, Bolton CH, Harvey RF. Changes in certain blood constituents during Ramadan. Am J Clin Nutr 1982; 36: 350–3.
7. Almond D, Mazumder B. Health capital and the prenatal environment: The effect of Ramadan observance during pregnancy. Am Econ J Appl Econ 2011; 3: 56–85.
8. Hood WW, Wilson CS. The literature of bibliometrics, scientometrics, and informetrics. Scientometrics 2001; 52: 291–314.
9. Pritchard A. Statistical bibliography or bibliometrics? J Doc 1969; 25: 348–9.
10. Beshyah WS, Beshyah SA. Bibliometric analysis of the literature on Ramadan fasting and diabetes in the past three decades (1989–2018). Diabetes Res Clin Pract 2019; 151: 313–22.
11. Husain S, Zafar M, Ullah R. Ramadan and public health: A bibliometric analysis of top cited articles from 2004 to 2019. J Infect Public Health 2020; 13: 275–80.
12. Kendall S. LibGuides: PubMed, Web of Science, or Google Scholar? A behind-the-scenes guide for life scientists. : Which one is best: PubMed, Web of Science, or Google Scholar? [Internet]. [cited 2020 Mar 15]. Available from: https://libguides.lib.msu.edu/c.php?g=96972&p=627295
13. Trusted publisher-independent citation database - Web of Science Group [Internet]. [cited 2020 May 29]. Available from: https://clarivate.com/webofsciencegroup/solutions/web-of-science/

Original Article

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Loss of smell in COVID-19 patients: is it related to clinical-radiological disease severity?

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ABSTRACT

Objective: Olfactory dysfunction (OD) is one of the most prominent predictive symptoms in the early detection of severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) disease (COVID-19), it may be the first symptom or accompany other symptoms. The predictive value of OD is unknown in terms of the overall prognosis of COVID-19. We aimed to investigate the relationship between OD and the clinical-radiological severity of the disease.

Material and Method: Data of 208 COVID-19 patients (105 inpatients and 103 outpatients) who had positive Real-Time Polymerase Chain Reaction (PCR) tests between December 1, 2020, and January 15, 2021, were collected retrospectively. Presence of OD, symptoms on admission other than OD, days of hospital stay, peripheral blood analysis values, COVID-19 disease severity [World Health Organization (WHO) 2020 "Clinical management of COVID-19"] and radiologic classifications [Radiological Society of North America Expert Consensus Statement on Reporting (RSNA) Chest CT Findings Related to COVID-19] were retrospectively collected.

Results: Analysis of 208 patients revealed that there were 105 (50.48%) inpatients and 103 (49.52%) outpatients. Among 102 patients who had OD, 68 were outpatients and 34 were inpatients. It was determined that the patients with OD were mostly followed up on an outpatient basis, and they did not need hospitalization ($p<0.0001$). The mean of hospital stay of 34 inpatients with OD was 7.52±4.63 days, while the mean of hospital stay of 71 patients without OD was 12.53±8.92 days, and those with OD were found to need a shorter hospital stay ($p=0.001$) and no relation was found between disease severity and the duration of OD ($p=0.381$). There was no significant difference in disease severity in relation to OD in the inpatient group ($p=0.71$).

Conclusion: OD is one of the most common symptoms of COVID-19. In the patients with loss of smell, the need for hospitalization is less, and hospital stay is shorter; these findings indicate that the patients with OD may experience a milder disease. The presence of OD may be used as a useful predictor by clinicians for the severity of the COVID-19 course.

Keywords: COVID-19, loss of smell, disease severity

INTRODUCTION

Studies have indicated that several viruses can use the olfactory nerve as the shortest route to the CNS and reported that post-viral anosmia may be the result of epithelial damage and involvement of the central nervous system, but the exact pathogenesis is uncertain (1).

COVID-19 is an ongoing viral pandemic that can lead to respiratory infections ranging from mild upper respiratory tract infections to fatal pneumonia (2,3). Fever, cough, diarrhea, shortness of breath, and myalgia have been identified as the most common and characteristic symptoms of COVID-19, and loss of smell and taste has recently been reported as frequent symptoms by many researchers (4). In February 2020, Mao et al. were the first authors who reported incipient olfactory or taste dysfunction as the symptoms of COVID-19 in addition to the known symptoms of the infection (5).

In our study, the relationship of the loss of smell with the clinical and radiological severity was investigated in COVID-19 patients.

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MATERIAL AND METHOD

This study was designed to obtain data cross-sectional and retrospectively. Institutional Education Board reviewed and approved the protocol of this retrospective study (Approval date December 24, 2020; Decree no:706). Also, the ethics committee approval for this study was obtained by Health Science University Ankara Keçiören Education and Training Hospital Clinical Researches Ethics Committee (Date: 28/12/2021, Decision No: 2012-KAEK-15/2439). All procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki.

Demographic, clinical, and radiological data of 208 PCR (+) COVID-19 patients (105 inpatients and 103 outpatients) between December 1, 2020, and January 15, 2021, were collected retrospectively. The OD was noted. The patients with loss of smell were contacted later by phone, to obtain information. Those who previously had olfactory problems were excluded from the study.

The laboratory results were recorded, and COVID-19 clinical severity was grouped as mild, moderate, severe, or critical according to the WHO 2020 guideline (Clinical management of COVID-19). Radiographic classification on HRCT and CXR grading of lung involvement were recorded according to RSNA Chest CT/ CXR Findings Related to COVID-19.

Statistical analysis: Categorical data were analyzed with the Chi-square test, and expressed as total number and percentage. Kolmogorov-Smirnov or Shapiro-Wilk test, coefficient of variation, skewness-kurtosis values, histogram, and detrended plot graphs were examined for the distribution of all continuous variables. Normally distributed variables were analyzed with the Student t-test or ANOVA, and non-normally distributed variables were analyzed with Mann-Whitney U and Kruskal-Wallis tests and presented as mean±SD and median (min-max), respectively. The relationship of hospitalization with other categorical data was analyzed with the Chi-square test. SPSS (Statistical Package for the Social Sciences) statistical software program (version 22) was used for statistical analysis. The significance level was set at $p \leq 0.05$.

RESULTS

A total of 208 PCR (+) COVID-19 patients were analyzed. The mean age (SD) was 52.3±14.8 years, 62% of them were males. There was a loss of smell in 59.5% of women and 42.6% of men. It was found that loss of smell was more pronounced in females ($p<0.018$).

Loss of smell was observed in 49% of all patients, 32% of inpatients, and 66% of outpatients, and there was a significant difference among the groups ($p<0.0001$). Loss of smell was the first symptom in 6.3% of all patients, and the difference between inpatients (4.8%) and outpatients (7.8%) was not significant ($p=0.37$) (**Table 1**).

Table 1. The demographic and clinical characteristics of the patients

	Total (n=208) (100%)	Inpatients (n=105) (50.48%)	Outpatients (n=103) (49.52%)	p-value
Age, mean±SD	52.32±14.84	59.23±14.06	45.27±12.07	p<0.0001
Gender				p=0.163
Male	129 (62%)	70 (66.7%)	59 (57.3%)	
Female	79 (38%)	35 (33.3%)	44 (42.7%)	
Smoking				p=0.001
Never	122(58.7%)	58 (55.2%)	64 (62.1%)	
Ex-smoker	53 (25.5 %)	37 (35.2%)	16 (15.5%)	
Current	33 (15.9%)	10 (9.5%)	23 (22.3%)	
Loss of smell				p<0.0001
Present	102 (49 %)	34 (32.4%)	68 (66%)	
Absent	106 (51%)	71 (67.6%)	35 (34%)	
The first symptom is loss of smell				p=0.371
Yes	13 (6.3%)	5 (4.8%)	8 (7.8%)	
No	195 (93.7%)	100 (95.2%)	95 (92.2%)	
Loss of smell in the first 5 days				p<0.0001
Yes	83 (39.9%)	27 (25.7%)	56 (54.4%)	
No	125 (60.1%)	78 (74.3%)	47 (45.6%)	
Fever				p<0.0001
Present	112 (53.8%)	71 (67.6%)	41 (39.8%)	
Absent	96 (46.2%)	34 (32.4%)	62 (60.2%)	
Cough				p=0.001
Present	90 (43.3%)	57 (54.3%)	33 (32%)	
Absent	118 (56.7%)	48 (45.7%)	70 (68%)	
Dyspnea				p<0.0001
Present	64 (30.8%)	52 (49.5%)	12 (11.7%)	
Absent	144 (69.2%)	53 (50.5%)	91 (88.3%)	
Throat ache				p=0.013
Present	42 (20.2%)	14 (13.3%)	28 (27.2%)	
Absent	166 (79.8%)	91 (96.7%)	75 (72.8%)	
Headache				p=0.237
Present	55 (26.4%)	24 (22.9%)	31 (30.1%)	
Absent	153 (73.6%)	81 (77.1%)	72 (69.9%)	
Myalgia				p=0.809
Present	145 (69.7%)	74 (70.5%)	71 (68.9%)	
Absent	63 (30.3%)	31 (29.5%)	32 (31.1%)	
Diarrhea				p=0.798
Present	29 (13.9%)	14 (13.3%)	15 (14.6%)	
Absent	179 (86.1%)	91 (86.7%)	88 (85.4%)	
Malaise				p=0.212
Present	126 (60.6%)	68 (64.8%)	58 (56.3%)	
Absent	82 (39.4%)	37 (35.2%)	45 (43.7%)	
Nausea				p=0.049
Present	13 (6.3%)	10 (9.5%)	3 (2.9%)	
Absent	195 (93.8%)	95 (90.5%)	100 (97.1%)	

Regarding COVID-19 severity; 32% of all patients had mild disease, and outpatients (63.1%) had a milder disease compared to inpatients (1.9%) ($p<0.0001$). None of the outpatients had severe or critical diseases (**Table 2**).

CXR was normal in 54.4% of outpatients and 15.2% of inpatients. Moderate and severe pneumonia (41% and 19%, respectively) were significantly more in hospitalized patients (p<0.0001). 43.7% of the outpatients and 5.7% of the hospitalized patients did not need further imaging with thorax CT. The typical radiological appearance of COVID-19 on thorax CT (GGO) was evident in 47.1 of all patients, 72.4% of inpatients, and 21.4% of outpatients, and the difference was significant (p<0.0001) (**Table 2**).

Table 2. The comparisons of the disease severity, CXR / HRCT radiological findings

	Total (n=208) (100%)	Inpatients (n=105) (50.48%)	Outpatients (n=103) (49.52%)	p-value
Clinical stage				p<0.0001
Asymptomatic	0	0	0	
Mild	67 (32.2%)	2 (1.9%)	65 (63.1%)	
Moderate	93 (44.7%)	55 (52.4%)	38 (36.9%)	
Severe	22 (13.9%)	29 (27.6%)	0 (0%)	
Critical	19 (9.1%)	19 (18.1%)	0 (0%)	
CXR findings				p<0.001
Normal	72 (34.6%)	16 (15.2%)	56 (54.4%)	
Mild pneumonia	57 (27.4%)	26 (24.8%)	31 (30.1%)	
Moderate pneumonia	59 (28.4%)	43 (41%)	16 (15.5%)	
Severe pneumonia	20 (9.6%)	20 (19%)	0 (0%)	
Thorax HRCT findings				p<0.001
Typical	98 (47.1%)	76 (72.4%)	22 (21.4%)	
Indeterminate	9 (4.3%)	5 (4.8%)	4 (3.9%)	
Atypical	16 (7.7%)	8 (7.6%)	8 (7.8%)	
Negative	34 (16.3%)	10 (9.5%)	24 (23.3%)	
HRCT not available	51 (24.5%)	6 (5.7%)	45 (43.7%)	

Comparisons of the groups that patients who had a loss of smell within the first 5 days after onset of the disease and after 5 days did not yield any differences between the groups in terms of disease severity (mild/moderate/severe/critical) classification and CXR pneumonia findings (normal/mild/moderate/severe) (not presented in the table) (p=0.513 and p=0.512; respectively). The analysis of disease severity between the hospitalized patients with OD (n=34) and those without OD (n=71) revealed that there was no significant difference between them (p=0.071).

The laboratory findings are presented in **Table 3**. D-dimer, ferritin, CRP, LDH, BUN, and AST levels were higher in hospitalized patients in which OD was observed at a lower rate (p<0.0001).

DISCUSSION

Many underlying disorders may result in OD. The most common causes are sinonasal disorders, upper respiratory tract infections, and head trauma. Inflammation, sinusitis, rhinitis, nasal polyps, and mucus may obstruct the nasal airway and block the olfactory area in the nose and are the most common causes of OD (50-70%) (6,7).

Coronaviruses have been identified as a family of viruses that may be associated with OD. Suzuki et al. showed coronaviruses in the nasal secretions of patients with OD. The authors demonstrated the virus antigen 60-66 hours after infection, and the viruses were mostly detected in the olfactory bulb, the pathophysiological mechanisms leading to OD in COVID-19 are still unknown (8,9).

Table 3. The comparison of laboratory findings between two groups

		Total (n=208) (100%) Mean±SD	Inpatients (n=105) (50.48%) Mean±SD	Outpatients (n=103) (49.52%) Mean±SD	p-value
WBC	$\times10^3/mm^3$	7.38 +3.37	8.15 ±3.94	6.59±2.46	p=0.001
Neutrophil	$\times10^3/mm^3$	5.31±3.10	6.39±3.57	4.20 ±2.03	p<0.0001
Lymphocyte	$\times10^3/mm^3$	1.55±1.04	1.29 ±1.04	1.82±0.98	p<0.0001
NLR		5.12±5.80	7.27 ±7.12	2.93 ±2.67	p<0.0001
Eosinophil	$\times10^3/mm^3$	0.056 ±0.073	0.029±0.054	0.085±0.079	p<0.0001
Hemoglobin	g/dl	13.84 ±2.04	13.63±1.63	14.05±2.37	p=0.131
Trombocyte	$\times10^3/mm^3$	249.85±96.16	258.63±115.94	240.89±69.98	p=0.184
D-dimer	mg/L	1.01±3.02	1.53±4.17	0.47±0.46	p=0.011
CRP	mg/L	47.02±65.74	81.91±74.93	11.46±22.69	p<0.0001
Troponin	ng/L	4.12±11.65	5.32±15.84	2.89±4.04	p=0.133
Ferritin	ng/ml	260.60±335.77	416.24±407.87	101.93±93.91	p<0.0001
LDH	IU/L	257.89±133.74	316.23±161.17	197.80±51.62	p<0.0001
Albumin	g/L	37.65±7.24	33.41±7.02	41.98±4.33	p<0.0001
BUN	mg/dL	16.13±7.66	19.71±8.76	12.48±3.76	p<0.0001
ALT	IU/L	31.93±27.31	35.20±31.71	28.61±21.59	p=0.082
AST	IU/L	33.05±32.36	39.25±42.39	26.72±14.58	p=0.005

Studies have reported that OD affected women significantly more (10). In our study, loss of smell was found in 59.5% of women and 42.6% of men, and it was determined that women were affected more in terms of OD ($p<0.018$). This finding was in line with previous studies. In addition, in the inpatient group, the rate of hospitalization of women (33.3%) was lower than that of men ($p<0.05$).

It was observed that the patients who did not need hospitalization were younger than the hospitalized patients ($p<0.05$). Loss of smell was observed in 32.4% of inpatients and 66% of outpatients, and the outpatients were younger. These results were consistent with the results of previous studies (11,12).

The prevalence of loss of smell was found to be higher in outpatients, however when all patients were taken into consideration, the rate was similar to the rate in Klopfenstein et al.'s study (47%) (13) but lower than the rates reported by Mao et al. and Lechien et al. (5,14).

Loss of smell appeared as the first symptom in 6.3% of our patients, and this rate was lower than the rate reported by Lechien et al. (11.8%) (14). A comparison of inpatients and outpatients for loss of smell did not yield any statistically significant difference. In our study, the median duration of loss of smell was found as 10 (1-60) days, which was similar to Klopfenstein et al.'s (13). study (8.9±6.3 [1–21]).

Similar to previous reports (15), 39% of our patients had a loss of smell in the early phase of COVID-19 (first 5 days), and the difference between outpatients and inpatients was statistically significant (54.4%vs. 25.7%) ($p<0.0001$).

Fever (53.8%), malaise (60.6%), and myalgia(69.7%) were the most frequent symptoms in our patients, similar to previous studies (16,17). Compared to outpatients, fever (67.6%), cough (54.3%) and dyspnea (49.9%) were significantly more frequent in our hospitalized patients ($p<0.0001$); other symptoms were not significantly different in the inpatient and outpatient groups.

COVID-19 severity (mild, moderate, severe, critical) was determined according to the WHO 2020 "Clinical management of COVID-19" algorithm. It was found that 32% of our whole patient population had mild disease, and 63.1% of our outpatients had milder disease compared to inpatients. Severe disease was seen in 13.9%, and critical disease was seen in 9.1% of our patients. Most of the cases were classified as mild (81%) in similar studies (severe patients 14%, critically ill patients 5%) (18).

In a study, it was reported that 58.3% of the patients had normal chest X-rays (19). In our study, CXR was evaluated as normal in 54.4% of outpatients, and 15.2% of inpatients. In addition, it was observed that clinicians did not request thoracic CT in 43.7% of outpatients (5.7% for inpatients). In the outpatient group who had a high rate of loss of smell, further imaging was not needed as the disease was mild and the CXR findings were normal or displayed mild pneumonia.

Typical ground-glass opacities (GGO) on thorax CT have been reported as the most frequent finding of COVID-19 by many authors. In our study, a typical GGO pattern was observed in 72.4% of the inpatient group, and in 21.4% of the outpatient group who had a loss of smell more frequently, with a significant difference in between ($p=0.001$).

Similar to other viral infections, lymphopenia is observed in COVID-19 patients (20). Some studies suggested that lymphopenia could be used as a prognostic factor for COVID-19. Zhao et al (21). showed that lymphopenia increased the risk of severe COVID-19 nearly threefold. Lower lymphocyte counts were reported in patients' central nervous system (CNS) symptoms compared to the ones without CNS symptoms, and the patients with severe infections had higher D-dimer levels compared to the ones with non-severe infections (22). Zhang et al. observed a relation between eosinopenia and lymphopenia in the prognosis of severe COVID-19 patients (23).

In our study, lymphocyte and eosinophil values were lower than outpatients in hospitalized patients who had a low rate of OD and high disease severity ($p=0.0001$). Neutrophil to lymphocyte ratio (NLR) is of great importance in showing the general inflammatory status (24), and studies showed higher NLR in severe COVID-19 patients (25).

In our study, WBC, neutrophil, and NLR values were higher in the inpatient group with a low rate of OD but with more severe disease and signs of pneumonia ($p=0.0001$). As a result that is not compatible with the existing studies in the literature; In our study, no significant difference was found between inpatient and outpatient groups in terms of Hb ($p=0.131$) and thrombocyte ($p=0.184$) levels (26,27).

High D-dimer level is a very frequent laboratory finding in COVID-19 patients. Guan et al. studied 1099 hospitalized COVID-19 patients and found high D-dimer (≥0.5 mg/L) as an indicator of severe disease. Tang et al. reported D-dimer as a predictor of mortality. Based on these findings, D-dimer has become a reliable indicator of prognosis, hospital mortality, and need for ICU care (17,28). In our study, the D-dimer level was approximately 3 times higher than the outpatients in the inpatient group with high disease severity.

Among biochemical markers, CRP, ferritin, procalcitonin, and LDH were shown to predict a poor prognosis. Ruan et al. reported a significant increase in CRP and serum ferritin levels (29). Similar to previous studies, we found CRP levels 7 times higher in the inpatient group with high disease severity compared to the outpatients ($p<0.0001$).

LDH ($p<0.0001$), ferritin ($p<0.0001$), BUN ($p<0.0001$), and AST ($p=0.005$) were significantly higher in the hospitalized patients compared to the outpatients. Albumin was lower in the hospitalized patients compared to the outpatients ($p<0.0001$), and this finding was consistent with the literature (29,30). There was no significant difference in troponin or ALT levels, and this was not in accordance with the literature (31,32).

Limitations: We did not employ tests of olfaction since this is a retrospective study, and the data were collected from the hospital's electronic recording system. In addition, information about the duration of OD is subject to recall bias.

CONCLUSION

In patients with loss of smell, the severity of COVID-19 disease is milder, CXR is often evaluated as normal, and further examinations such as thorax CT are less needed. Laboratory values, which have prognostic importance in the COVID-19 disease process, show parallelism with the severity of the disease (low lymphocyte, eosinophil values, high CRP, D-dimer values) in patients with low olfactory loss symptoms. Loss of smell is one of the primary symptoms to be questioned in COVID-19 patients. The presence of olfactory loss may assist the clinician in predicting prognosis based on symptoms.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Health Science University Ankara Keçiören Education and Training Hospital Clinical Researches Ethics Committee (Date: 28/12/2021, Decision No: 2012-KAEK-15/2439).

Informed Consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

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REFERENCES

1. van Riel D, Verdijk R, Kuiken T. The olfactory nerve: a shortcut for influenza and other viral diseases into the central nervous system. J Pathol 2015; 235: 277-87.
2. Su S, Wong G, Shi W, et al. Epidemiology, Genetic Recombination, and Pathogenesis of Coronaviruses. Trends Microbiol 2016; 24: 490-502.
3. Yıldız M, Özturk Ergur F, Uzel Senel M, Özturk A. The use of prone positioning in severe COVID-19 outside the intensive care unit. Bratisl Lek Listy 2021; 122: 590-3.
4. Bagheri SH, Asghari A, Farhadi M, et al. Coincidence of COVID-19 epidemic and olfactory dysfunction outbreak in Iran. Med J Islam Repub Iran 2020; 34: 62.
5. Mao L, Jin H, Wang M, et al. Neurologic Manifestations of Hospitalized Patients With Coronavirus Disease 2019 in Wuhan, China. JAMA Neurol 2020; 77: 683-90.
6. Damm M, Temmel A, Welge-Lüssen A, et al. Epidemiologie und Therapie in Deutschland, Osterreich und der Schweiz [Olfactory dysfunctions. Epidemiology and therapy in Germany, Austria and Switzerland]. HNO 2004; 52: 112-20. German.
7. Li X, Lui F. Anosmia. [Updated 2021 Sep 25]. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2022 Jan-. Available from: https: //www.ncbi.nlm.nih.gov/books/NBK482152/
8. Suzuki M, Saito K, Min WP, et al. Identification of viruses in patients with postviral olfactory dysfunction. Laryngoscope 2007; 117: 272-7.
9. Netland J, Meyerholz DK, Moore S, Cassell M, Perlman S. Severe acute respiratory syndrome coronavirus infection causes neuronal death in the absence of encephalitis in mice transgenic for human ACE2. J Virol 2008; 82: 7264-75.
10. Lefèvre N, Corazza F, Valsamis J, et al. The Number of X Chromosomes Influences Inflammatory Cytokine Production Following Toll-Like Receptor Stimulation. Front Immunol 2019; 10: 1052.
11. Gandhi RT, Lynch JB, Del Rio C. Mild or moderate COVID-19. N Engl J Med 2020; 383: 1757-66.
12. Yan CH, Faraji F, Prajapati DP, Ostrander BT, DeConde AS. Self-reported olfactory loss associates with outpatient clinical course in COVID-19. Int Forum Allergy Rhinol 2020; 10: 821-31.
13. Klopfenstein T, Kadiane-Oussou NJ, Toko L, et al. Features of anosmia in COVID-19. Med Mal Infect 2020; 50: 436-9.
14. Lechien JR, Chiesa-Estomba CM, De Siati DR, et al. Olfactory and gustatory dysfunctions as a clinical presentation of mild-to-moderate forms of the coronavirus disease (COVID-19): a multicenter European study. Eur Arch Otorhinolaryngol 2020 Aug; 277: 2251-61.
15. Meini S, Suardi LR, Busoni M, Roberts AT, Fortini A. Olfactory and gustatory dysfunctions in 100 patients hospitalized for COVID-19: sex differences and recovery time in real-life. Eur Arch Otorhinolaryngol 2020; 277: 3519-23.
16. Wang D, Hu B, Hu C, et al. Clinical Characteristics of 138 Hospitalized Patients With 2019 Novel Coronavirus-Infected Pneumonia in Wuhan, China. JAMA 2020; 323: 1061-9.
17. Huang C, Wang Y, Li X, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. Lancet 2020; 395: 497-506.

18. Wu Z, McGoogan JM. Characteristics of and Important Lessons From the Coronavirus Disease 2019 (COVID-19) Outbreak in China: Summary of a Report of 72 314 Cases From the Chinese Center for Disease Control and Prevention. JAMA 2020; 323: 1239-42.
19. Weinstock MB, Echenique A, Russell JW, et al. Chest x-ray findings in 636 ambulatory patients with COVID-19 presenting to an urgent care center: a normal chest x-ray is no guarantee. J Urgent Care Med 2020; 14: 13-18.
20. Zhang D, Guo R, Lei L, et al. Frontline Science: COVID-19 infection induces readily detectable morphologic and inflammation-related phenotypic changes in peripheral blood monocytes. J Leukoc Biol 2021; 109: 13-22.
21. Zhao Q, Meng M, Kumar R, et al. Lymphopenia is associated with severe coronavirus disease 2019 (COVID-19) infections: A systemic review and meta-analysis. Int J Infect Dis 2020; 96: 131-5.
22. Mao L, Jin H, Wang M, et al. Neurologic manifestations of hospitalized patients with coronavirus disease 2019 in Wuhan, China. JAMA Neurol 2020 1; 77: 683-90.
23. Zhang JJ, Dong X, Cao YY, et al. Clinical characteristics of 140 patients infected with SARS-CoV-2 in Wuhan, China. Allergy 2020; 75: 1730-41.
24. Liu Y, Du X, Chen J, et al. Neutrophil-to-lymphocyte ratio as an independent risk factor for mortality in hospitalized patients with COVID-19. J Infect 2020; 81: 6-12.
25. Kong J, Wang T, Di Z, et al. Analysis of hematological indexes of COVID-19 patients from fever clinics in Suzhou, China. Int J Lab Hematol 2020; 42: 204-6.
26. Omrani-Nava V, Maleki I, Ahmadi A, Moosazadeh M, et al. Evaluation of Hepatic Enzymes Changes and Association with Prognosis in COVID-19 Patients. Hepat Mon 2020; 20: e103179.
27. Yang X, Yang Q, Wang Y, et al. Thrombocytopenia and its association with mortality in patients with COVID-19. J Thromb Haemost 2020; 18: 1469.
28. Tang N, Li D, Wang X, Sun Z. Abnormal coagulation parameters are associated with poor prognosis in patients with novel coronavirus pneumonia. J Thromb Haemost 2020; 18: 844-7.
29. Ruan Q, Yang K, Wang W, Jiang L, Song J. Clinical predictors of mortality due to COVID-19 based on an analysis of data of 150 patients from Wuhan, China. Intensive Care Med 2020; 46: 846-8.
30. Liu Y, Yang Y, Zhang C, et al. Clinical and biochemical indexes from 2019-nCoV infected patients linked to viral loads and lung injury. Sci China Life Sci 2020; 63: 364-74.
31. Mishra AK, Lal A, Sahu KK, Sargent J. Cardiovascular factors predicting poor outcome in COVID-19 patients. Cardiovasc Pathol 2020; 49: 107246.
32. Mao R, Qiu Y, He JS, et al. Manifestations and prognosis of gastrointestinal and liver involvement in patients with COVID-19: a systematic review and meta-analysis. Lancet Gastroenterol Hepatol 2020; 5: 667-78.

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Mean platelet volume (MPV) levels in subclinical hypothyroidism and its relation to serum lipid levels

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ABSTRACT

Background: Subclinical hypothyroidism (SCH) is a term to define a mild and early thyroid deficiency. Some reports claim SCH has an association with increased cardiovascular disease (CVD), especially in individuals <70 years old. Mean platelet volume (MPV) is thought to be an independent marker for CVD. Dyslipidemia is also a common entity in thyroid related disorders and a tradational biomarker of increased risk of CVD. This study aims to reveal the MPV levels in SCH and to discuss its relation to lipid profile.

Material and Method: This retrospective single-center study was conducted between 2009 March and August. 94 newly diagnosed patients with SCH and 79 controller were enrolled in the study. Patients with normal serum levels of free T3 and T4 and >4 mIU/L TSH levels were labelled for SCH group. Both groups were evaluated for MPV, lipid profiles and epidemiological features.

Results: SCH group had higher levels of MPV, total cholesterol (TC) and LDL cholesterol (LDL-C) levels (P<0.05, for each parameter). MPV had a positive correlation to TC and LDL-C (r=0,175 ve p=0,025; r=0,154 and p=0,043). However, a multivariate analysis that rendered the impact of age and gender revealed that SCH had no impact on MPV after adjustment for those parameters gender revealed that SCH had no impact on MPV after adjustment for those parameters (B=0,304; 95% CI: -0,021 – 0,628; p=0,066).

Conclusion: SCH patients may have higher levels of MPV. However, age and gender had an impact on MPV levels and we think increased CVD risk in SCH patients is associated with factors other than MPV levels.

Keywords: MPV, lipids, subclinical hypothyroidism

INTRODUCTION

Subclinical hypothyroidism (SCH) refers to an elevated serum thyrotropin (or as commonly known, thyroid stimulating hormone [TSH]) accompanying with normal levels of free thyroxine (fT4). SCH effects 3% to 8% of the general population in the world while some regions are more incident (1,2). A common approach for the treatment of SCH involves to treat individuals with TSH >10 mU/L or elevated TSH accompanying with symptoms that thought to be related with hypothyroidism (2).

SCH may have some adverse impact on blood pressure, lipid levels, and atherosclerosis which all are the traditional strong risk factors for the future cardiovascular disease (CVD) or events. So it is logical to think on whether the SCH is a modifiable factors for CVD (3-6). Since it has been shown dyslipidemia reversed in overt hypothyroidism patients following levothyroxyine therapy (6,7), it is a key question whether to treat SCH may provide some benefits in those patients.

Mean platelet volume (MPV) is one of predictor parameters of platelet function which has been linked to cardiovascular disease (8-9). Some studies claim SCH is associated with higher MPV levels (9). In contrast, others have not indicated an association between those two parameters in their studies with diverse cohorts (10-12). However all studies are designed on completely various protocols and many factors may have effect on results.

We aimed to test whether SCH is associated with MPV and MPV levels is related to lipid levels in our cohort to contribute the data pool of the existing literature.

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MATERIAL AND METHOD

This single-center retrospective study was conducted between March and August 2009 in Yıldırım Beyazid University Dışkapı Education and Training Hospital. Ninety-four individuals with subclinical hypothyroidism and seventy-nine healthy individuals (laboratory findings were normal and with a disease-free history) were enrolled in the study. Laboratory and clinical features of the participants were noted.

- **SCH** was defined as; serum TSH levels >4 mIU/L plus normal serum fT3 and fT4.
- **Groups**: Group 1; SCH and Group 2; healthy individuals
- **Exclusion**: Existence of those clinical features; overt hypo-hyperthyroidism or being under a treatment for a known thyroid disease, diabetes mellitus, long-standing hypertension, coronary artery disease, dyslipidemia or being under lipid-lowering therapy, hematological disease, chronic kidney disease and all other conditions that indicates an acute systemic disease.
- **Comparision parameters**: serum TSH, fT4, fT3, MPV, total cholesterol, low-density lipoprotein cholesterol (LDL-c), high-density cholesterol (HDL-c), fibrinogen, high sensitive c-reactive protein (hsCRP), fasting glucose, platelet counts, hemoglobin and hemotocrit.
- **Blood sampling**: All blood samples were drawn between 08:00 and 10:00 at morning and all samples were studied in the central laboratory of the hospital at the admission day.
- **Data collection**: Data were collected via central hospital automation software, noted by a authorised doctor, and all individuals were queried again for exclusion criteria by phone calls.

This study was produced from the specialization thesis. The study was carried out with the permission of Health Ministry of Turkish Republic Dışkapı Yıldırım Beyazıt Training and Research Hospital Ethical Commitee (Date: 13.12.2021, Decision No:126/03). All procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki.

Statistical Assessment

Dataset were analysed via using Statistical Package for the Social Sciences (S.P.S.S.) version 11.5 (IBM SPSS Corp. NY, USA). Normality of the parameters was assessed by performing the Kolmogorov-Smirnov test. Continuous parametric variables were presented as mean ± standard deviation and non-parametric variables as median with maximum and minimum levels. Categorical variables were expressed as percentiles (%), and qualitative measures were evaluated by using Chi-Square test. Continuous parametric and non-parametric variables were compared by independent Sample T test and Mann-Whitney U-test, respectively. Multivariate analysis were performed to describe the factors might have impact on MPV levels. A correlation between continuous parametric and non-parametric variables were analysed via Spearmen's and Pearson's correlation tests, respectively. P value <.05 was indicated as statistically significant. The power analysis of the data was calculated from the observed power of the parameters by considering Type b error (<0.2).

RESULTS

A total of 173 individuals were evaluated. Group 1 was older (41.9±14.9 vs 30.1±9.3) and included more female (female= 144 and male= 29) individuals compared to Group 2; p <0.001 and p= 0.019, respectively. Laboratory features of both two groups were given in **Table 1**.

Table 1. Comparison of the laboratory findings of the two groups

Variables	Group 1, n= 94	Group 2, n= 79	P value
Hgb, gr/dl	13.5±1.39	13.8±1.43	0.146
HTC, %	39.4±3.72	39.8±4.06	0.432
PLT, µg/L	275 (140-552)	248 (118-443)	<0.001
MPV,fl	8.5 (6.2-11.0)	8.1 (6.1-11.4)	0.004
Glucose, mg/d	92 (72-120)	81 (58-138)	<0.001
TSH, mIU/L	5.8 (4.1-30.1)	1.6 (0.3-3.9)	<0.001
fT4, ng/dl	1.2 (0.3-4.97)	1.3 (0.99-1.8)	<0.001
fT3, pg/ml	3.3 (1.6-55.0)	1.27 (1.02-1.93)	0.065
TC, mg/dL	190.5 (110-298)	162 (99-260)	<0.001
TG, mg/dL	122 (46-376)	98 (39-430)	0.041
LDL-c, mg/dL	115.1±36.39	94.6±31.80	<0.001
HDL-c, mg/dL	45.5 (25-91)	49 (5-84)	0.052
Fibrinogen, mg/dl	341.5 (184-910)	240 (120-370)	<0.001

Hgb: hemoglobin, Htc: hematocrit, Plt: platelet, MPV: mean platelet volüme, TSH: thyroid stimulanting hormone, fT4: free T4, fT3: free T3

Pearsons' correlation test revealed a positive correlation between MPV and TSH (p<0.001, r2= 0.079) (**Figure 1**). Then after we divided Group 1 according to TSH levels into three subgroups to determine whether higher levels of TSH are associated with higher levels of MPV (TSH >4 and <6 mIU/L, TSH ≥6 and <10 mIU/L, and TSH ≥10 mIU/L) (**Table 2**). However, those three groups were found similar for MPV in Anova test; p=0.898. Age was similar among three subgroups; p=0.078.

Table 2. Comparison of MPV, fibrinojen, TG, TC, LDL-c, and HDL-c in subgroups of SCH

Variables	TSH >4 and <6 mIU/L	≥6 and <10 mIU/L	≥10 mIU/L	p
Age, years	39.2±12.1	42.1±16.7	40.1±13.1	0.872
MPV, fl	8.5±0.98	8.4±1.06	8.6±0.93	0.898
Fibrinogen, mg/dl	353 (189-804)	304.5 (184-910)	340.0 (253-866)	0.117
TG, mg/dl	122 (55-296)	135 (46-376)	120 (64-281)	0.613
TC, mg/dl	186 (110-298)	202.5 (136-284)	196 (113-298)	0.622
LDL-c, mg/dl	112.2±37.57	117.3±38.07	120.0±32.49	0.685
HDL-c, mg/dl	45 (29-91)	47 (30-68)	45 (25-60)	0.958

TSH: thyroid-stimulating hormone, TG: triglyceride, TC: total cholesterol, LDL-c: low-density lipoprotein cholesterol, HDL-c: high-density lipoprotein cholesterol

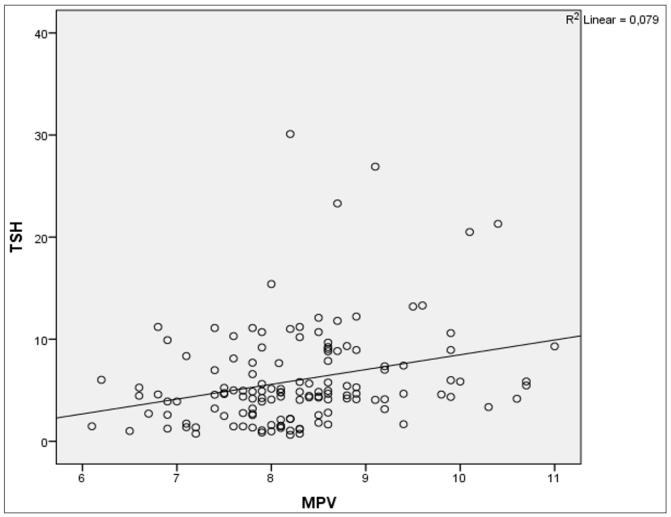

Figure 1. Corellation between TSH and MPV

Univariate analysis did not reveal a relation between MPV and age, TG, and HDL-c in Group 1 (p>0.05). MPV has also posed a positive correlation with TC and HDL-c (r= 0.175 and p= 0.025; r= 0.154 and p= 0.043, respectively).

A regression model was used to investigate the impact of SCH, age, TC, HDL-c and gender on MPV, and we found that SCH had no impact on MPV levels after adjustment (p= 0.06, B= 0.304, CI %95; -0.021 – 0.628) (**Table 3**) (however if CI would have been set up as 90%, p value could demonstrate a significant level, or if the study had a larger sample size result would be significant since p=0.06 was close to p=0.05). On the other hand, when the same model was applied to fibrinogen in Group 1, it was observed that SCH and age remained to have an impact on fibrinogen (p<0.001, B=0,334; 95% CI; 0,236 – 0,431) (**Table 4**).

Table 3. Multivariate analysis of the factors impact on MPV

Variables	Regresyon Coefficient (B)	p	%95 CI (B)	
			Lower	Upper
SCH	0.304	0.066	-0.021	0.628
Age	0.0002	0.980	-0.014	0.014
Male	-0.307	0.127	-0.701	0.088
TC	0.001	0.809	-0.007	0.009
TG	0.001	0.593	-0.002	0.003
HDL-c	-0.0002	0.966	-0.009	0.008

SCH: subclinical hypothyroidism, TC: total cholesterol, TG: triglyceride, HDL-c: high-density lipoprotein

Table 4. Multivariate analysis of the factors impact on fibrinogen

Variables	Regresyon Coefficient (B)	p	%95 CI (B)	
			Lower	Upper
SCH	0.334	<0.001	0.236	0.431
Age	0.004	0.038	0.0003	0.009
Male	-0.037	0.542	-0.155	0.082
TC	-0.001	0.616	-0.003	0.002
TG	-0.001	0.252	-0.001	0.0004
HDL-c	-0.002	0.516	-0.006	0.003

DISCUSSION

Subclinical hypothyroidism has dysmetabolic effects such as dyslipidemia, increased blood pressure and glucose which are all traditional cardiovascular risk factors (13). MPV is the average size of platelets found in blood and is thought to be an marker of platelet activity which is relavent to atherothrombotic events (14,15). Overt hypo/hyperthyroidism have been found to associated with increased MPV, however, impact of SCH on MPV is doubtful. Our clinical trial indicates SCH has no impact on MPV and is also not related to lipid profiles.

SCH is a common type of thyroid hormone disorder worldwide and has well-known dyscardiometabolic effects. Myocardial and vascular endothelial tissues are sensitive to changes in the concentrations of circulating thyroid hormones and adverse events may occur even in the case of SCH (16). However, the mechanism involved in the pathogenesis of SCH-related cardiovascular adverse events is still speculative (16,17). Moreover, guidelines recommend to treat the SCH individuals with cardiovascular disease due to probable benefits of to treat and evidence of adverse events related to not treated patients (17).

Thyroid hormone receptor is expressed by hematopoietic stem cells and a low/high concentration of thyroid hormone is associated with clonogenecity and apoptosis of hematopoietic system (18,19). Plateles are enucleated cells and produced by megakaryocytes in bone marrow, have a lifespan of 7-10 days. MPV is determined during megakaryocytogenesis and influenced by many factors which play key role in thrombopoiesis, as well as thyroid hormone (20). Additionally, thyroid hormones are associated with increased platelet levels, however, the mechanism is unknown (21). MPV is thought to be an activity marker of platelets so it is plausible to establish the strategies to determine the factors might result in increase of MPV. Impact of SCH on MPV is controversial and our study also has not determined a relation between MPV and SCH. Nevertheless, in our cohort we found a positive correlation between TSH and MPV levels considered all individuals of the cohort. Then after we tried to find a hazard range for TSH which would determine a risk for increased MPV. However, we could not find such a link between two parameters. This might be due to some other factors that have effects on MPV. Platelet levels were found higher in SCH group in our study.

SCH has been found associated with a higher level of blood glucose and dyslipidemia as expected, as demonstrated in previous studies. MPV levels have not had an association with lipid levels.

Our study has some limitations. Some factors which might have impact on MPV levels, such as smoking, blood pressure, body mass index, were not investigated due to the lack of data. Our cohort has a small sample size and only one hemogram and biochemical study results could have been evaluated. Despite all, we think our findings may contribute to the literature on the topic.

CONCLUSION

Our study reveals SCH have no impact on MPV. SCH might be contributing to cardiovascular adverse events via various pathways other than MPV levels.

ETHICAL DECLARATION

Ethics Committee Aproval: The study was carried out with the permission of Health Ministry of Turkish Republic Dışkapı Yıldırım Beyazıt Training and Research Hospital Ethical Commitee (Date: 13.12.2021, Decision No:126/03).

Informed Consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

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Author Contributions: The authors declare that they have all participated in the design, execution, and analysis of the article, and that they have approved the final version.

REFERENCES

1. Fatourechi V. Subclinical hypothyroidism: an update for primary care physicians. Mayo Clin Proc 2009; 84: 65-71.
2. Biondi B, Cappola AR, Cooper DS. Subclinical Hypothyroidism: A Review. JAMA 2019; 322: 153-60.
3. Suh S, Kim DK. Subclinical Hypothyroidism and Cardiovascular Disease. Endocrinol Metab (Seoul) 2015; 30: 246-51.
4. Liu D, Jiang F, Shan Z, et al. A cross-sectional survey of relationship between serum TSH level and blood pressure. J Hum Hypertens 2010; 24: 134-8.
5. Ye Y, Xie H, Zeng Y, Zhao X, Tian Z, Zhang S. Association between subclinical hypothyroidism and blood pressure-a meta-analysis of observational studies. Endocr Pract 2014; 20:150-8.
6. Razvi S, Ingoe L, Keeka G, Oates C, McMillan C, Weaver JU. The beneficial effect of L-thyroxine on cardiovascular risk factors, endothelial function, and quality of life in subclinical hypothyroidism: randomized, crossover trial. J Clin Endocrinol Metab 2007; 92: 1715-23.
7. Pearce EN. Update in lipid alterations in subclinical hypothyroidism. J Clin Endocrinol Metab 2012; 97: 326-33.
8. Yılmaz H, Ertuğrul Ö, Ertuğrul B, Ertuğrul D. Mean platelet volume in patients with subclinical hypothyroidism. Platelets 2011; 22: 143-7.
9. Kim JH, Park JH, Kim SY, Bae HY. The mean platelet volume is positively correlated with serum thyrotropin concentrations in a population of healthy subjects and subjects with unsuspected subclinical hypothyroidism. Thyroid 2013; 23: 31-7.
10. Kutluturk F, Gul SS, Sahin S, Tasliyurt T. Comparison of mean platelet volume, platelet count, neutrophil/ lymphocyte ratio and platelet/lymphocyte ratio in the euthyroid, overt hypothyroid and subclinical hyperthyroid phases of papillary thyroid carcinoma. Endocr Metab Immune Disord Drug Targets 2019; 19: 859-65.
11. Torun AN, Uzum AK, Aksoy N. Overt and mild subclinical hypothyroidism do not influence mean platelet volume in premenopausal women having low cardiac risk. Clin Appl Thromb Hemost 2012; 18: 312-5.
12. Ren X, Meng Z, Liu M, et al. No associations exist between mean platelet volume or platelet distribution width and thyroid function in Chinese. Medicine (Baltimore) 2016; 95:e4573.
13. Gao F, Wang G, Xu J. Alteration of hemostatic parameters in patients with different levels of subclinical hypothyroidism and the effect of l-thyroxine treatment. Ann Clin Lab Sci 2017; 47: 29-35.
14. Coban E, Kucuktag S, Basyigit S. Platelet activation in subjects with impaired glucose tolerance. Platelets 2007; 18: 591-4.
15. Park Y, Schoene N, Harris W. Mean platelet volume as an indicator of platelet activation: methodological issues. Platelets 2002; 13: 301-6.
16. Gasparyan AY, Ayvazyan L, Mikhailidis DP, Kitas GD. Mean platelet volume: a link between thrombosis and inflammation? Curr Pharm Des 2011; 17: 47-58.
17. Jabbar A, Pingitore A, Pearce SH, Zaman A, Iervasi G, Razvi S. Thyroid hormones and cardiovascular disease. Nat Rev Cardiol 2017 Jan;14: 39-55.
18. Cappola AR, Desai AS, Medici M, et al. Thyroid and cardiovascular disease: research agenda for enhancing knowledge, prevention, and treatment. Thyroid 2019; 29: 760-77.
19. Grymuła K, Paczkowska E, Dziedziejko V, et al. The influence of 3,3',5-triiodo-L-thyronine on human haematopoiesis. Cell Prolif 2007; 40: 302-15.
20. Chute JP, Ross JR, McDonnell DP. Minireview: Nuclear receptors, hematopoiesis, and stem cells. Mol Endocrinol 2010; 24: 1-10.
21. Josefsson EC, White MJ, Dowling MR, Kile BT. Platelet life span and apoptosis. Methods Mol Biol 2012; 788: 59-71.

Original Article

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Base excess, bicarbonate, and lactate levels predict 28-day mortality in patients with COVID-19: a retrospective study

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ABSTRACT

Aim: It is critical to categorize the risk factors that could disclose the severity of COVID-19. This study aimed to determine the effects of arterial blood gases on hospital mortality by examining the results retrospectively measured at the first admission to the emergency department of cases diagnosed with COVID-19.

Material and Method: In this retrospective study, arterial blood gases of patients with COVID-19 were analyzed using univariate analysis to identify factors linked to 28-day all-cause in-hospital mortality. The patients were divided into two groups survivors and nonsurvivors.

Results: The study included 159 survivors and 33 nonsurvivors with COVID-19. Serum levels of lactate, D-dimer, troponin, and CRP were higher and serum levels of base excess, bicarbonate, and albumin to creatinine ratio were lower in nonsurvivor patients than in survivors. The highest AUC was found for lactate and base excess.

Conclusion: The arterial blood gases performed during the first admission to the emergency department are linked with disease severity and can be used to predict disease progression and mortality. Furthermore, patients with higher levels of lactate and lower levels of base excess and bicarbonate should be monitored closely and treated early.

Keywords: COVID-19, base excess, bicarbonate, lactate, mortality

INTRODUCTION

Coronavirus disease (COVID-19) is a viral infection that presents with severe pneumonia and spreads rapidly to many countries as a pandemic (1). The viral agent of the disease was identified as a new coronavirus (2019-nCoV, SARS-CoV-2) (2). COVID-19 has infected over 250 million individuals and killed over 5 million of them (3). The symptoms of COVID-19 are fever, cough, dyspnea, myalgia, sore throat, and dyspnea (4). The clinical course of COVID-19 may differ between individuals. Especially in patients with comorbidities such as hypertension, diabetes, chronic obstructive pulmonary disease, deaths due to acute respiratory distress syndrome, septic shock, metabolic acidosis, disseminated intravascular coagulopathy and multiple organ failure may occur (5). One of the most delicate challenges in hospitals with a large number of patient admissions is evaluating critical patients. It is crucial to determine which people are at high risk of dying. It is critical to categorize the risk factors that could disclose the severity of COVID-19.

Although the gold-standard test in the diagnosis of the disease is polymerase chain reaction tests (PCR) performed with real-time reverse transcriptases, changes occur in biochemical tests such as urea, creatinine, CRP, ferritin, D-dimer, troponin, lymphocyte and neutrophil levels due to the inflammatory process caused by the virus (6,7). The biochemical tests used routinely provide clinicians with important data about COVID-19. Many studies have been conducted to date on the effect of biochemical parameters on mortality in COVID-19 patients, and some risk factors have been found, such as comorbid diseases, D-dimer, and troponin (8-12). However, the number of publications evaluating the effect of arterial blood gases such as lactate, base excess, and bicarbonate on mortality is very few and the clinical consequences of these results during the first admission remain unclear. We aimed to determine the effects of arterial blood gases and biochemical tests on hospital mortality by examining the results retrospectively measured at the first admission to the emergency department (ED) of cases diagnosed with COVID-19.

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MATERIAL AND METHOD

This study was reviewed and approved by the Human Research Ethical Board of Muğla Sıtkı Koçman University (Date: 24/03/2021, Decision No: 44). This study was performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments.

Study Design

This retrospective, observational study was designed to investigate the relationship between biochemical parameters and 28-day all-cause in-hospital mortality in adult patients with COVID-19 who were referred to the ED of Muğla Training and Research Hospital and hospitalized between March 2020 and February 2021. Diagnoses of COVID-19 were made in accordance with WHO guidelines (13) and confirmed by real-time reverse transcriptase-PCR (RT-PCR) assay. Patients were excluded from the study if they had a diagnosis of pregnancy, anemia, cancer, hematologic or rheumatologic diseases, readmissions, or were aged <18 years. Demographic information, radiological reports, comorbidities, and laboratory results were obtained from the hospital information system and then correlated with clinical outcomes.

Patient Evaluation

After being admitted to the ED, these patients were assessed by an emergency medicine physician as a standard procedure in our hospital. Routine biochemical tests, RT-PCR test, arterial blood gases, and chest computed tomography (CT) of the patients were requested. Patients who underwent a consultant's evaluation were admitted based on the results of laboratory tests and CT scans. Patients were admitted to clinics or the intensive care unit (ICU) depending on the severity of COVID-19.

The radiological diagnosis of COVID-19 was recorded using the "CO-RADS classification," a reporting system for COVID-19, which ranged from 1 (very unlikely) to 5 (very likely) (14). CO-RADS scores of 1–2 were defined as incompatible with COVID-19, a score of 3 was suspicious for COVID-19, and scores of 4–5 were compatible with COVID-19.

Statistical analysis

The Statistical Package for Social Sciences was used to analyze the data (Version 22.0, SPSS Inc., Chicago, IL). The Kolmogorov–Smirnov test was used to determine the normality of the quantitative data distribution. Nonparametric tests (such as the Mann–Whitney U–test and the Kruskal–Wallis test) were used on nonnormally distributed data, whereas parametric tests (such as the independent samples t-test and Tukey's post hoc test) were used on normally distributed data. Summary statistics were expressed as the mean, standard deviation, or median (minimum-maximum). Statistical significance was determined for those differences with a p-value of 0.05 or below. Receiver operating characteristic (ROC) curve analysis was used to assess diagnostic accuracy. The area under the ROC curve (AUC) was used to determine the accuracy of these tests. Higher AUC values indicate better test performance. A parameter with an AUC value equal to 1 discriminates individuals perfectly as survivors or nonsurvivors. The Kaplan–Meier test was used to compute the cumulative survival rate, and the log-rank test was used to examine differences in survival across the groups. The data were first analyzed using univariate analysis to identify factors linked to in-hospital mortality. Next, significant factors were employed in a stepwise forward logistic regression analysis. Additionally, sensitivity and specificity analyses for mortality were carried out.

RESULTS

After collecting the medical data, 192 consecutive patients presenting to the ED were selected for further study from a total of 291 records. The exclusions of patients were shown in the flowchart (**Figure 1**). The patients with COVID-19 were divided into two groups who were discharged from the hospital (survivors, n=159) and died in the hospital in 28-day (nonsurvivors, n=33). Nonsurvivors had a median survival time of 8 days (range, 1-28 days). Post hoc power calculations were applied, and the sample size was seen to provide 0.999 power and 1.007 effect size for lactate levels at an α error probability level of 0.05. The mean age of survivors was 55.4±17.5 (range 19-92 years), while the mean age of nonsurvivors was 70.2±13.9 (range 34-99 years). There were age and gender differences between the groups. Nonsurvivors were predominantly male (64%; p <0.001) and older than the survivors (70.2 vs 55.4 years, p <0.001). The baseline characteristics of the patients are shown in **Table 1**.

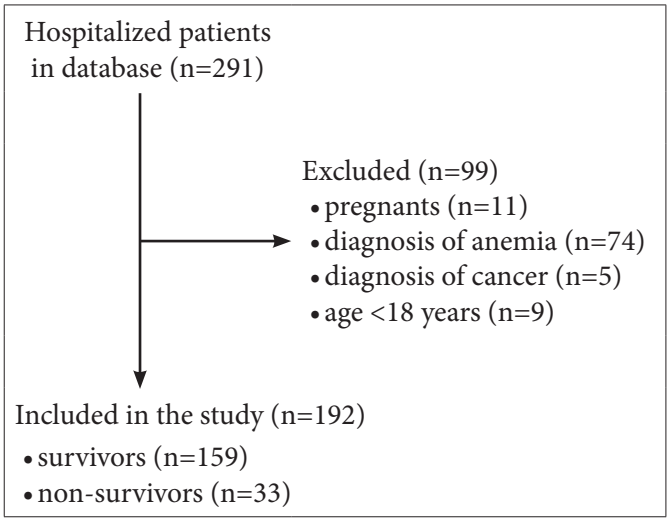

Figure 1. Flowchart of the study

Table 1. Baseline characteristics of the groups according to in-hospital mortality

Variables	Survivors (n=159)	Non-survivors (n=33)	p value
Age (years)	55.4±17.5	70.3±13.9	<0.001
Gender (Women/Men)	78/81	12/21	<0.001
Comorbid disease	52 (32.7%)	30 (90.9%)	<0.001
Hypertension	26 (16.3%)	20 (60.6%)	<0.001
Diabetes mellitus	24 (15.9%)	12 (36.3%)	0.007
Coronary artery disease	15 (9.4%)	7 (21.2%)	0.071
Chronic obstructive pulmonary disease	12 (7.5%)	10 (30.3%)	0.001
Chronic renal failure	2 (1.25%)	7 (21.2%)	<0.001
CT findings			
Incompatible with COVID-19	41 (25.7%)	1 (3.0%)	<0.001
Suspicious for COVID-19	9 (6.8%)	0 (0.0%)	<0.001
Compatible with COVID-19	109 (68.5%)	32 (96.9%)	<0.001
Hospitalization			
Ward	134 (84.3%)	1 (3%)	<0.001
Intensive care unit	25 (15.7%)	32 (97%)	<0.001
Biochemical parameters			
White blood cell (×103/μL)	5.82 (2.36, 25.27)	10.15 (2.44, 23.36)	<0.001
Lymphocyte (×103/μL)	1.34 (0.22, 3.53)	0.61 (0.12, 2.80)	<0.001
Neutrophil (×103/μL)	3.59 (1.00, 21.95)	8.37 (1.91, 20.15)	<0.001
CRP (mg/L)	50.43 (0.60, 361.47)	108.27 (1.57, 364.61)	<0.001
D-Dimer (ng/mL)	880.77 (42, 8651)	2643.54 (142, 8405)	<0.001
Troponin (pg/mL)	9.67 (3, 120)	52.82 (3, 401)	<0.001
Albumin (g/L)	40.74±5.05	31.83±7.58	<0.001
Creatinine (mg/dL)	0.96 (0.48, 5.91)	1.92 (0.36, 7.65)	<0.001
Alb/Cr	43.80±13.93	29.76±16.84	<0.001
Blood gas parameters (arterial)			
pH	7.41±0.07	7.39±0.11	0.616
Base excess (mmol/L)	1.74 (-24.9, 10.6)	-4.32 (-17.3, 4.7)	0.007
Bicarbonate (mmol/L)	27.3±3.8	24.1±6.7	0.003
Lactate (mmol/L)	1.2 (0.4, 15.0)	1.6 (0.5, 12.3)	0.012

Summary statistics are given as the mean ± standart deviation, median (interquartile ranges), and count (percentages). CRP: C reactive protein, Alb/Cr: albumin to creatinine ratio.

A total of 82 (42.7%) patients had comorbidities. The most prevalent comorbidities were hypertension (24.0%) and diabetes mellitus (18.8%). In addition, a history of hypertension, diabetes mellitus, chronic obstructive pulmonary disease, and chronic renal failure was associated with high mortality (p<0.001, p=0.007, p=0.001, and p<0.001, respectively). However, there was no statistically significant difference in the history of coronary artery disease. CT findings that were compatible with COVID-19 were observed in 141 (73.4%) patients. Fifty-seven patients (29.7%) were hospitalized in the ICU and 135 (70.3%) patients were followed in clinics. Thirty-three patients (17.2%) died in the hospital during 28 days.

The levels of base excess and bicarbonate during the first admission to ED were significantly lower and lactate levels were higher in nonsurvivors compared with survivors (p=0.007, p=0.003, p=0.012, respectively). In terms of the laboratory results, there were statistically significant differences in CRP, D-dimer, troponin, and albumin to creatinine ratio between the groups (**Table 1**, all p-values <0.001).

The areas under the ROC curves (AUCs) were 0.892, 0.842, 0.793, 0.749, 0.741, 0.725, and 0.723 for lactate, base excess, bicarbonate, troponin, D-dimer, albumin to creatinine ratio, and CRP, respectively, for in-hospital mortality (**Figure 2**). The sensitivity, specificity, and accuracy rates for in-hospital mortality based on lactate of 2.0 mmol/L were 84.8%, 81.1%, and 81.7%, respectively (95% CI, AUC: 0.892, p <0.001). The sensitivity, specificity, and accuracy rates for in-hospital mortality based on a base excess of 0.1 mmol/L were 78.7%, 84.2%, and 83.3%, respectively (95% CI, AUC: 0.842, p <0.001). The sensitivity, specificity, and accuracy rates for in-hospital mortality based on bicarbonate of 20.0 mmol/L were 75.7%, 94.9%, and 91.6%, respectively (95% CI, AUC: 0.793, p <0.001).

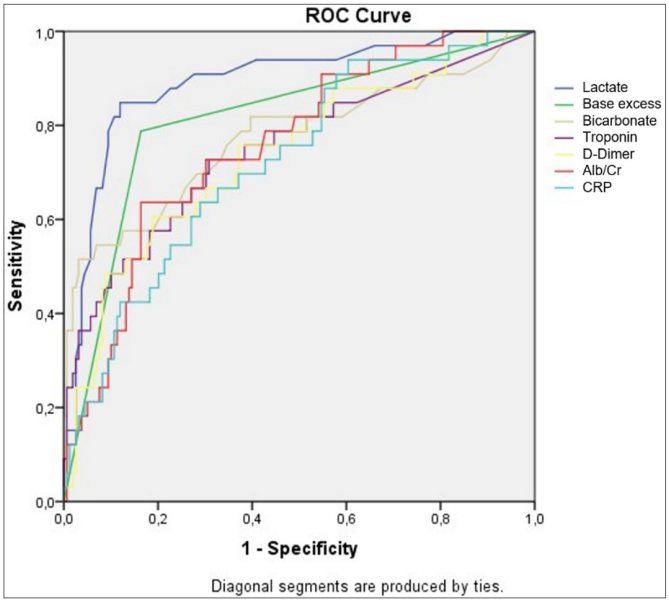

Figure 2. ROC curve for lactate (0.892), base excess (0.842), bicarbonate (0.793), troponin (0.749), D-dimer (0.741), albumin to creatinine ratio (0.725), and CRP (0.723)

A total of 33 patients died in hospital. Twenty-eight of these patients had a lactate level ≥ 2.0 mmol/L, 26 patients had a base excess ≤ 0.1 mmol/L and 25 patients had a bicarbonate ≤ 20.0 mmol/L (Fisher's exact test, all p values <0.001). **Figure 3a**, **Figure 3b**, and **Figure 3c** show the Kaplan–Meier survival curves for lactate, base excess, and bicarbonate according to these cutoff values. Patients with lactate levels above the cutoff value had significantly higher in-hospital mortality rates than those with levels below the cutoff value according to Kaplan–Meier curves (log-rank test=2.663; p <0.001).

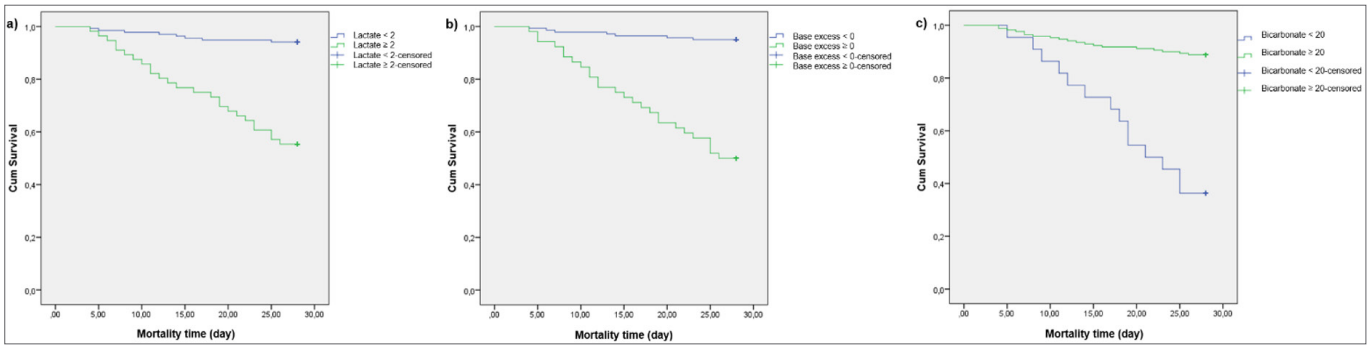

Figure 3. Kaplan–Meier survival curve with (a) lactate ≥ 2 mmol/L, (b) base excess ≤ 0.1 mmol/L, and (c) bicarbonate ≤ 20 mmol/L

Base excess, comorbid disease, lactate, bicarbonate, hypertension, D-dimer, troponin, CRP, Alb/Cr, and age were independent predictors of in-hospital mortality with odds ratios of 12.894, 11.705, 9.368, 7.394, 6.218, 5.213, 4.234, 3.283, 1.149, and 1.048, respectively (Cox regression test, all p values <0.001; **Table 2**).

Table 2. Cox regression analysis for the prediction of in-hospital mortality

Variables	Odds ratio	95% CI	p value
Base excess	12.894	5.584-29.773	<0.001
Comorbid disease	11.705	4.112-33.322	<0.001
Lactate	9.368	4.219-20.802	<0.001
Bicarbonate	7.394	3.917-18.741	<0.001
Hypertension	6.218	3.087-12.524	<0.001
D-Dimer	5.213	2.590-10.493	<0.001
Troponin	4.234	2.105-8.516	<0.001
CRP	3.283	1.562-6.901	0.002
Alb/Cr	1.149	1.083-1.219	<0.001
Age	1.048	1.025-1.071	<0.001

CRP: C reactive protein, Alb/Cr: albumin to creatinine ratio

DISCUSSION

In this study, arterial blood gases and biochemical tests were evaluated as mortality risk factors in patients with COVID-19 during the first admission to the ED. The most significant finding of the study was that base excess, lactate, bicarbonate, hypertension, D-dimer, troponin, CRP, Alb/Cr, and age were independent mortality predictors in patients.

With respect to demographic factors such as age and sex, Dong et al. (15) divided patients with COVID-19 into two groups, severe and mild, and found that severe cases were significantly older with a greater proportion of males. In our study, nonsurvivor patients were prevalently male and older than survivor patients which were consistent with findings in the literature that older men are more susceptible.

Comorbidities in patients with COVID-19 are another significant mortality predictor as described in previous meta-analysis studies (16,17). In a study by Guan et al. (18), the presence of any coexisting illness was more common among patients with severe disease than among those with the nonsevere disease. In our study, 82 (42.7%) of the patients had comorbidities, with hypertension (24.0%) being the most common. We observed that comorbidities were independent predictors of mortality similar to previous studies (OR: 11.705, p<0.001).

Arterial blood gases are reliable tests to evaluate mortality in COVID-19, as described in previous studies (19-22). In a study by Bruno et al. (19), a decreasing lactate concentration over time was inversely associated with ICU mortality. Another study by Dheir et al. (20) showed higher levels of pH and lactate in patients who died. Kieninger et al. (21) identified blood pH value, mean arterial pressure, base excess, troponin, and procalcitonin as highly significant prognostic factors of in-hospital mortality. However, no significant difference was found for other parameters expected to be relevant prognostic factors, such as high lactate levels. This conflicting result of lactate may be attributed to the low sample size of the study. Kunt et al. (22) studied a very large range of parameters in nonsurvivor patients. They found higher levels of lactate, D-dimer, fibrinogen, CRP, and troponin and lower levels of partial pressure of carbon dioxide, base excess, and bicarbonate in patients who died in the hospital. In our study, higher serum levels of lactate, D-dimer, troponin, and CRP and lower levels of base excess, bicarbonate, and albumin to creatinine ratio were associated with in-hospital mortality in accordance with the literature. Cox-regression analysis represented higher odds ratios for base excess and lactate levels than those studies (odds ratios for BE: 12.894 and lactate: 9.368, respectively, p<0.001).

Previous studies have demonstrated biochemical parameters as relevant prognostic factors in COVID-19 (11,23,24). Bonetti et al. (24) found higher levels of CRP, D-dimer, and troponin in patients who died in the hospital and concluded that these parameters are highly predictive of in-hospital death and may be useful for guiding risk assessment and clinical decision-making. Pan et al. (11) also found that sex, SpO2, breath rate,

diastolic pressure, neutrophils, lymphocytes, CRP, procalcitonin, lactate dehydrogenase, and D-dimer were significantly correlated with death events. In the current study, CRP, D-dimer, and troponin levels were found to be significantly higher in the nonsurvivor group than in the survivor group, in line with these findings. To the best of our knowledge, this is the first study to have evaluated the albumin to creatinine ratio in predicting COVID-19 mortality, and the results showed that lower levels of Alb/Cr were found in nonsurvivor patients. Renal and liver pathologies should have driven this decrease.

The main strength of this study was the well-designed comparison of biochemical parameters in patients with COVID-19 during the first admissions to the ED. However, there were some limitations to the study. Primarily, our study design was retrospective, single-center, and had limited data. Second, this was a cross-sectional study, which cannot describe the cause and effect relationship between laboratory parameters and clinical outcomes. Nevertheless, despite these limitations, this study can be considered valuable with respect to mortality predictors and the determination of cutoff values and odds ratios between arterial blood gases in a comparison of survivors and nonsurvivors.

CONCLUSION

Arterial blood gases performed during the first admission to the ED are linked with disease severity. Blood levels of base excess, lactate, and bicarbonate can be used to predict disease progression and mortality. Furthermore, patients with older age, male sex, hypertension, higher levels of lactate, D-dimer, troponin, CRP, and lower levels of base excess, bicarbonate, and albumin to creatinine ratio should be monitored closely and treated early. These parameters have the potential to give frontline clinicians better triage of COVID-19 patients in the ED. Overall, our findings can be useful in clinical practice and should be confirmed by large-scale studies.

ETHICAL DECLARATIONS

Ethics Committee Approval: This study was carried out with the permission of Muğla Sıtkı Koçman University, Human Research Ethics Committee (Date: 24/03/2021, Decision No: 44).

Informed consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

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Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper and that they have approved the final version.

REFERENCES

1. Lu H, Stratton CW, Tang YW. Outbreak of pneumonia of unknown etiology in Wuhan, China: The mystery and the miracle. J Med Virol 2020; 924: 401-2.
2. Zhu N, Zhang D, Wang W, et al. A Novel Coronavirus from Patients with Pneumonia in China, 2019. N Engl J Med 2020; 3828: 727-33.
3. WHO Coronavirus Disease (COVID-19) Dashboard. Access address: https://covid19.who.int/ Access date: 06/08/2021.
4. Huang C, Wang Y, Li X, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. The Lancet 2020; 39510223: 497-506.
5. Paules CI, Marston HD, Fauci AS. Coronavirus Infections-More Than Just the Common Cold. Jama 2020; 323: 707–8.
6. Corman VM, Landt O, Kaiser M, et al. Detection of 2019 novel coronavirus (2019-nCoV) by real-time RT-PCR. Euro Surveill 2020; 253: 2000045.
7. Liu Y, Yang Y, Zhang C, et al. Clinical and biochemical indexes from 2019-nCoV infected patients linked to viral loads and lung injury. Sci China Life Sci 2020; 633: 364-74.
8. Bezuidenhout MC, Wiese OJ, Moodley D, et al. Correlating arterial blood gas, acid-base and blood pressure abnormalities with outcomes in COVID-19 intensive care patients. Ann Clin Biochem 2021; 582: 95-101.
9. Güven BB, Ertürk T, Yıldız E, Durmayüksel E, Ersoy A, Tanoğlu A. Our convalescent plasma experiences in COVID-19 patients hospitalized in the intensive care unit. J Health Sci Med 2022; 5: 600-6.
10. Güngörer B. Baseline demographic, clinical and laboratory risk factors for predicting admission to intensive care unit in patients diagnosed with COVID-19 in the emergency department. Anatolian Curr Med J 2021; 3; 279-83.
11. Kara U, Şimşek F, Özhan MÖ, et al. The factor analysis approach to mortality prediction in COVID-19 severe disease using laboratory values: a retrospective study. J Health Sci Med 2022; 5: 528-33.
12. Bennouar S, Bachir Cherif A, Kessira A, et al. Usefulness of biological markers in the early prediction of corona virus disease-2019 severity. Scand J Clin Lab Invest 2020; 808: 611-8.
13. Diagnosis and Treatment Protocol for Novel Coronavirus Pneumonia (Trial Version 7). Chinese Med J 2020; 1339: 1087-95.
14. Prokop M, Everdingen Wv, Vellinga TvR, et al. CO-RADS: a categorical CT assessment scheme for patients suspected of having COVID-19—definition and evaluation. Radiology 2020; 2962: E97-E104.
15. Dong Y, Zhou H, Li M, et al. A novel simple scoring model for predicting severity of patients with SARS-CoV-2 infection. Transbound Emerg Dis 2020; 676: 2823-9.
16. Izcovich A, Ragusa MA, Tortosa F, et al. Prognostic factors for severity and mortality in patients infected with COVID-19: A systematic review. PLoS One 2020; 1511: e0241955.

17. Yang L, Jin J, Luo W, Gan Y, Chen B, Li W. Risk factors for predicting mortality of COVID-19 patients: A systematic review and meta-analysis. PLoS One 2020; 1511: e0243124.
18. Guan W-j, Ni Z-y, Hu Y, et al. Clinical Characteristics of Coronavirus Disease 2019 in China. New England Journal of Medicine 2020; 38218: 1708-20.
19. Bruno RR, Wernly B, Flaatten H, et al. Lactate is associated with mortality in very old intensive care patients suffering from COVID-19: results from an international observational study of 2860 patients. Ann Intensive Care 2021; 111: 128.
20. Dheir H, Karacan A, Sipahi S, et al. Correlation between venous blood gas indices and radiological involvements of COVID-19 patients at first admission to emergency department. Rev Assoc Med Bras (1992) 2021; 67: 51-6.
21. Kieninger M, Sinning A, Vadász T, et al. Lower blood pH as a strong prognostic factor for fatal outcomes in critically ill COVID-19 patients at an intensive care unit: A multivariable analysis. PLoS One 2021; 169: e0258018.
22. Türkay Kunt A, Kozaci N, Torun E. Mortality predictors in patients diagnosed with COVID-19 in the emergency department: ECG, laboratory and CT. Medicina (Kaunas) 2021; 57: 629.
23. Asghar MS, Haider Kazmi SJ, Khan NA, et al. Poor prognostic biochemical markers predicting fatalities caused by COVID-19: a retrospective observational study from a developing country. Cureus 2020; 128: e9575.
24. Bonetti G, Manelli F, Patroni A, et al. Laboratory predictors of death from coronavirus disease 2019 (COVID-19) in the area of Valcamonica, Italy. Clin Chem Lab Med 2020; 587: 1100-5.

Original Article

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Cardiac effects of Sugammadex and Rocuronium combination in rats: experimental study

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ABSTRACT

Aim: In this experimental study, it was aimed to examine the effects of rocuronium and sugammadex complex on cardiac muscle cells in rats histopathologically and biochemically.

Matherial and Method: 32 adult Sprague-Dawley male rats were divided into four groups with 8 in each group. Group 1 consisted of animals that did not undergo surgical treatment. Group 2 received a volume equivalent to 16 mg/kg sugammadex with 0.9% intravenous saline. Group 3 received 16 mg/kg of intravenous sugammadex. Group 4, 1 mg/kg intravenous rocuronium and 16 mg/kg intravenous sugammadex were administered to rats. After the procedure completed GSH and MDA level evaluated biochemically; and heart tissue evaluated histopathologically.

Results: In group 4, connective tissue edema between muscle fibers was observed to be significantly increased, vessel dilatation and hemorrhagic areas were observed. Groups 3 and 4 were found to cause an increase in GSH level when compared to Groups 1 and 2, and a decrease in MDA level in these two groups compared to the others.

Conclusion: Although sugammadex and sugammadex-rocuronium complex cause biochemical and histopathological effect on the heart tissue, there were no irreversible histopathologic changes and no significant biochemical difference found in this study.

Keywords: Sugammadex, rocuronium, cardiac muscle, histopathology, rat

INTRODUCTION

Neuromuscular blockers are often used to facilitate endotracheal intubation and improve surgical comfort (1). Neuromuscular blockers (NMB) are divided into two groups as depolarizing and non-depolarizing. Rocuronium is a non-depolarizing neuromuscular blocking agent frequently used in clinical practice. To shorten the recovery period, to regain muscle functions and to avoid postoperative pulmonary complications; the neuromuscular blockade created by the NMB agent, needs to be reversed (2). Widely used anticholinesterase agents (including neostigmine) increase the amount of acetylcholine (Ach) at the neuromuscular junction, thereby eliminating the effect of NMB agents by competitive inhibition. Sugammadex, on the other hand, is a modified gamma-cyclodextrin molecule and terminates its effects by encapsulating steroid NMB agents (3).

Many studies conducted since 2008, when it was approved for use in Europe, have proven the efficacy and safety of sugammadex (4-7). According to the results of recent meta-analysis studies, sugammadex used after neuromuscular block provides faster recovery compared to neostigmine (4-6).

Since sugammadex provides rapid and safe recovery, its clinical use is increasing day by day. Although many studies have emphasized that sugammadex is effective and safe, some studies have revealed that it has potential risks (8).

In this experimental study, it was aimed to evaluate the effects of sugammadex on cardiac tissue in rats by histopathological and biochemical methods.

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MATERIAL AND METHOD

Ethical Statement

The study protocol and permissions were reviewed and approved by the Animal Experiments Local Ethics Committee of Adıyaman University Faculty of Medicine (Decision No: ADYU-HADYEK: 2018-11/2). All procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki.

Animals and Experimental Design

A standard diet and water were given to Sprague-Dawley rats. The temperature and humidity of the environment were monitored daily and kept constant with 12-hour light-dark cycles. The animals were divided into four groups:

Group 1: consisted of animals that did not undergo surgical treatment.

Group 2: Rats were administered 0.9% intravenous saline with a volume equivalent to 16 mg/kg sugammadex.

Group 3: Rats received 16 mg/kg intravenous sugammadex (Bridion®; Schering - Plow Corporation, Oss, The Netherlands).

Group 4: Rats were administered 1 mg/kg intravenous rocuronium (Esmeron®; Organon, Istanbul, Turkey) and 16 mg/kg intravenous sugammadex (Bridion®; Schering -Plow Corporation, Oss, The Netherlands) three minutes later.

Sample size: Sample size was calculated based on MDA measurement in reference article with G-Power 3.1 program 80% power and 95% confidence (9). Effect size was found 1.45 and 8 rats were needed for each group. 32 Sprague-Dawley rats was planned for 4 groups in this study.

All drugs were administered intravenously to the tail vein. After drug administration, the rats were followed up with ECG (electrocardiography) for 3 days. After the procedure. All animals were euthanized and the heart tissues of the rats were removed under anesthesia.

Histopathological Evaluation

When the experimental application procedures were completed, the heart tissue samples of the groups were taken and fixed in 10% formaldehyde solution for 1 week. After the fixation of the tissues was completed, routine histological tissue follow-up consisting of alcohol, xylene and paraplast chemicals was performed. Tissue samples were then made into paraffin blocks. Thin sections of 5 µm thickness were taken from paraffin blocks for histopathological examination. The prepared sections were deparaffinized using xylene. Stained with Hematoxylin-Eosin (H&E) and Masson triple staining method. The stained sections were examined with a Carl Zeiss brand Axiocam ERc5 model digital camera attached microscope and histopathologically evaluated. Histopathological changes were evaluated in 2 groups as reversible and irreversible (10) (**Table 1**).

Table 1. Reversible-irreversible cell injury

Reversible cell injury	Irreversible cell injury
Cell swelling	Necrosis
Membrane blebs	Fibrosis
Fatty changes	Apoptosis

Biochemical Evaluation

The excised heart tissue samples were washed with saline at +4°C, stored in eppendorf tubes according to cold chain principles and at -70°C until analysis. In tissue samples, homogenates were cold prepared with a 0.15 M KCl (10%, w/v) homogenizer for tissue malondialdehyde (MDA) and glutathione peroxidase (GSH-Px) measurements.

Tissue MDA concentration was prepared according to the Uchiyama method (11). Tissue MDA concentrations were measured below 532 nm in nmol/g tissue.

GSH-Px analysis was performed according to the method described by Ellman (12). It was measured with a spectrophotometer at a wavelength of 410 nm.

Statistical Analysis

One-way analysis of variance (ANOVA) test was used in the analysis of malondialdehyde (MDA) and glutathione peroxidase (GSH-Px) values of the groups. The Mann-Whitney U test was used to compare the groups. Kruskal wallis test was used for data that did not show normal distribution, and Dunnet test was used for multiple comparisons.

RESULTS

In the follow-up period after drug administration, neuromuscular block returned in all rats administered sugammdex after rocuronium in group 4. None of rats was died during procedure. No side effects were observed associated with sugammadex administration (QT prolongation, bradycardia, allergy). In histopathologic examination, no changes was observed in group 1 and group 2. Interstitial edema and vessel dilatation were observed in group 3. Connective tissue edema, vessel dilatation and haemorrhagic areas were observed in group 4. Fibrosis and necrosis was not observed in any group. There was no significant difference found in MDA and GSH levels between groups.

Histopathological evaluation of the groups as a result of histological examination of the sections belonging to Group 1, no pathological findings were found. A structure consisting of dense muscle fibers with normal morphological structure and core placement of the fibers was dominant (**Figure 1. a1** and **a2**). In addition, in the triple staining findings, dilatation, connective tissue fibrosis and inflammation were not found in the vascular structures. In addition, no connective tissue edema was observed between the muscle fibers (**Figure 1 a3**).

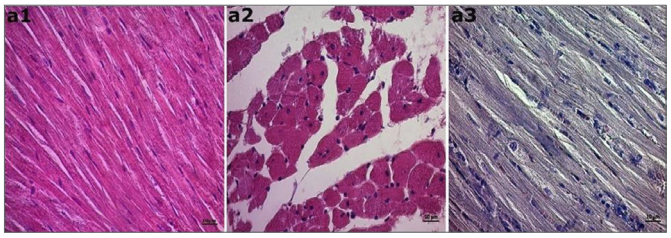

Figure 1. Light microscopy image of group 1; (**a1** and **a2** x40 objective magnification, H&E staining) (**a3** x40 objective magnification, Masson trichrome staining) Healthy tissue image

As a result of histological examination of the sections of group 2, a similar tissue image was found with group 1. There were dense muscle fibers and the connective tissue ratio between muscle fibers was normal (**Figure 2 b1** and **b2**). As a result of the evaluation made with triple staining, no signs of hemorrhage, connective tissue edema, inflammation and fibrosis were found. A healthy tissue image was observed (**Figure 2 b3**).

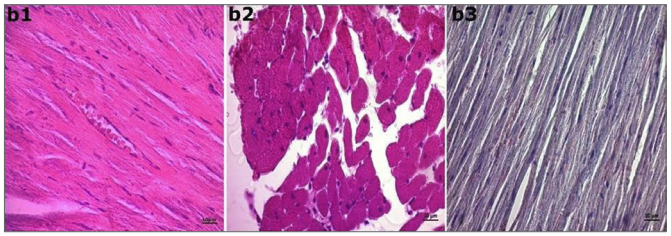

Figure 2. Light microscopy image of group 2; (**b1** and **b2** x40 objective magnification, H&E staining) (**b3** x40 objective magnification, Masson trichrome staining) Group 1 and similar healthy tissue image

As a result of the histological examination of the sections of group 3, there was a slight increase in the number of fibers with interstitial edema and morphological changes in places compared to groups 1 and 2. In addition, signs of vessel dilatation were observed (**Figure 3 c1**, **c2** and **c3**). As a result of the evaluation made with triple staining, no signs of inflammation and fibrosis were found (**Figure 3 c4**).

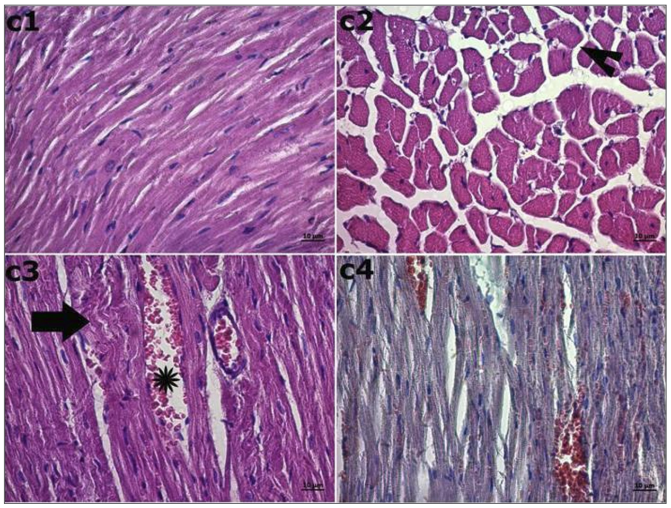

Figure 3. Light microscopy image of group 3; (c1, c2 and c3 x40 objective magnification, H&E staining) (c4 x40 objective magnification, Masson trichrome staining) Black arrow; distorted muscle fiber, star; dilated vessel, black arrowhead; interstitial edema between muscle fibers.

As a result of histological examination of the sections belonging to Group 4, degeneration of muscle fibers was the most intense group compared to other groups. Connective tissue edema between muscle fibers was markedly increased. There was vessel dilatation (**Figure 4 d1, d2** and **d3**). As a result of the evaluation made with triple staining, no signs of inflammation and fibrosis were found. However, haemorrhagic areas were observed (**Figure 4 d4**).

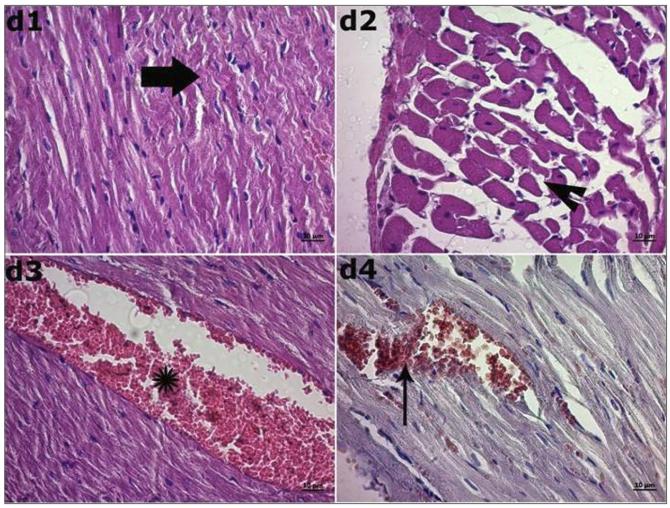

Figure 4. Light microscopy image of group 4; (d1, d2 and d3 x40 objective magnification, H&E staining) (d4 x40 objective magnification, Masson trichrome staining) Black arrow; distorted muscle fiber, star; dilated vessel, black arrowhead; interstitial edema between muscle fibers; thin black arrow; hemorrhagic area.

When the GSH level, a parameter belonging to the antioxidant systems, was compared between the groups, it was determined that group 3 and group 4 increased ($p>0.005$). There was a decrease in MDA level, which is an indicator of lipid peroxidation, in these two groups compared to the others. These increases and decreases were not found to be statistically significant (**Table 2**).

Table 2. Tissue GSH and MDA values

Groups	Statistics	GSH nmol/g	MDA nmol/g
Group 1	Mean	8964.4	288.5
	Median (Min-Max)	9163.1 (4410.1-12403.4)	283.1 (244.6 – 338.4)
Group 2	Mean	7042.2	267.2
	Median (Min-Max)	7262.5 (4628-10172.7)	271.4 (241.2-298.2)
Group 3	Mean	9280.9	259.6
	Median (Min-Max)	9865 (5756.2 -12243.1)	259.6 (227.8-288.1)
Group 4	Mean	9963.5	260.5
	Median (Min-Max)	9874.6 (4487-14871.2)	249.6 (211.1-328.3)
	p	0.272	0.175

Groups 3 and 4 caused an increase in GSH level compared to Groups 1 and 2. We believe that these substances cause an acute increase in GSH level due to the oxidative system. MDA levels did not cause a level of toxicity that

could cause a significant difference between the groups. Sugammadex and rocuronium did not cause oxidative stress-induced lipid peroxidation. The insignificant increase in MDA level suggests that these substances do not show severe toxicity as a result of free radical-induced oxidative stress and lipid peroxidation in heart tissue.

DISCUSSION

Sugammadex has no effect on NM connectivity or any receptor system in the body. This eliminates the need for anticholinergic drugs, which are alternatives and have many side effects. Clinical studies have shown that the side effects of sugammadex are mild and short-lived.

Side effects of sugammadex on the cardiovascular system have been reported as prolongation of the QT interval, bradycardia, hypotension, rhythm disturbances, and in rare cases asystole (13-17).

Sugammadex; although the drug was approved for use in Europe in 2008, the American Food and Drug Administration (FDA) postponed the approval for use until the end of 2015 due to the risk of serious hypersensitivity reactions. In their study, Tsur and Kalansky (18) identified 15 possible hypersensitivity reactions worldwide.

Considering the clinical studies examining the effects of sugammadex on the cardiac system; although high rates of bradycardia are mentioned in some publications, the incidence of bradycardia in patients treated with sugammadex was reported as 2% in a recent meta-analysis study (4).

In studies examining the relationship between sugammadex and QT prolongation, no significant correlation was found even in high-dose sugammadex use (19,20).

Dahl et al. (19) reported that sugammdex-rocuronium complex does not have a muscarinic effect due to its inert structure; therefore, it had no significant effect on hemodynamics and QT interval

Considering the experimental studies, Bostan et al. (21) reported histopathological changes in renal tissue without deterioration of biochemical renal function values in rats administered rocuronium and high-dose sugammadex (96mg/kg). In a similar study, they reported that rocuronium-sugammadex complex caused edema and degeneration in the heart and diaphragm muscle tissues. It has been suggested that the effects may not be directly related to sugammadex, and that the aminosteroid structure of rocuronium may have an effect on myopathy (22). In our study, tissue edema, vessel dilatation and haemorrhagic areas were detected in the histopathological examination of the sugammadex-rocuronium group, but no signs of inflammation or fibrosis were found.

In an I/R study examining the effects of rocuronium-sugammadex complex on brain tissue; neither biochemical values (GSH, MDA) nor histopathologically significant changes were found (9). In our study, no significant difference was found between the groups in terms of GSH and MDA levels.

Considering the recent ischemia-reperfusion (I/R) studies; there are studies in the literature reporting that sugammadex has organ-protective effects. In a recent I/R study, renal ischemia was performed and the nephroprotective effect of high-dose (100mg/kg) sugammadex use was demonstrated (23). In an I/R study performed with bilateral carotid occlusion for 10 minutes, sugammadex was reported to be protective against cerebral ischemia (24).

In this study, the dose of sugammadex that is used safely in humans was used. Although higher doses have been used in some studies, a dose of 16mg/kg has been used in many studies (9,21,22). There was no significant difference found in cardiac and diaphragmatic skeletal muscle with 16 mg/kg and 96 mg/kg sugammadex doses (22). Because of this study aimed to predict the side effects of sugammadex in clinical use, the study was planned to be conducted with this dose.

we observed that sugammadex and rocuronium-sugammadex complex made histopathological changes on the heart muscle in an experimental model in rats. Inthe group given rocuronium and sugammadex, the degeneration of the muscle fibers as a result of the histological examination of the sections was the most intense group compared to the other groups. Connective tissue edema between muscle fibers was markedly increased. Vascular dilatation was present. But no haemorrhagic areas were observed. In the group given sugammadex, less than the group given sugammadex-rocuronium complex, there was an increase in the number of interstitial edema and fibers with morphological changes from place to place. In addition, vascular dilatation findings were observed.

The GSH levels in our experimental groups were found to be higher than those in the control group, we think that this is an acute increase caused by the oxidative system. MDA levels did not cause a level of toxicity that could cause a significant difference between the groups. Sugammadex and rocuronium did not cause oxidative stress-induced lipid peroxidation. The insignificant increase in MDA level suggests that these substances do not show severe toxicity as a result of free radical-induced oxidative stress and lipid peroxidation in heart tissue.

CONCLUSION

As a result, in this experimental study, in parallel with the clinical studies performed, sugammadex and sugammadex-rocuronium complex cause biochemical and histopathological effect on the heart tissue, but no irreversible changes (fibrosis,necrosis) was found in histopathologic evaluation and no significant difference was found in MDA and GSH levels that used as an index for oxidative cellular damage.

ETHICAL DECLARATIONS

Ethics Committee Approval: This study was approved by Animal Experiments Local Ethics Committee of Adıyaman University Faculty of Medicine (Decision No: ADYU-HADYEK: 2018-11/2).

Informed Consent: Because of experimental design of the study informed consent form was not obtained

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

Financial Disclosure: The authors declared that this study has received no financial support.

Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

REFERENCES

1. Fisher DM. Clinical pharmacology of neuromuscular blocking agents. Am J Health Syst Pharm 1999; 56: 4–9.
2. de Boer HD, van Egmond J, van de Pol F, Bom A, Booij LHDJ. Chemical encapsulation of rocuronium by synthetic cyclodextrin derivatives: Reversal of neuromuscular block in anaesthetized Rhesus monkeys. Br J Anaesth 2006; 96: 201-6.
3. Bom A, Bradley M, Cameron K, et al. A novel concept of reversing neuromuscular block: chemi-cal encapsulation of rocuronium bromide by a cyclodextrin-based synthetic host. Angew Chem Int Ed Engl 2002; 41: 266-70.
4. Carron M, Zarantonello F, Tellaroli P, Ori C. Efficacy and safety of sugammadex compared to neostigmine for reversal of neuromuscular blockade: a meta-analysis of randomized controlled trials. J Clin Anesth 2016; 35: 1-12.
5. Hristovska AM, Duch P, Allingstrup M, Afshari A. The comparative efficacy and safety of sugammadex and neostigmine in reversing neuromuscular blockade in adults. A Cochrane systematic review with meta-analysis and trial sequential analysis. Anaesthesia 2018; 73: 631-41.
6. Abad-Gurumeta A, Ripollés-Melchor J, Casans-Francés R, et al.A systematic review of sugammadex vs neostigmine for reversal of neuromuscular blockade. Anaesthesia 2015; 70: 1441-52.
7. Won YJ, Lim BG, Lee DK, Kim H, Kong MH, Lee IO. Sugammadex for reversal of rocuronium-induced neuromuscular blockade in pediatric patients: a systematic review and meta-analysis. Medicine (Baltimore) 2016; 95: e4678.
8. Lee W. The potential risks of sugammadex. Anesth Pain Med 2019; 14: 117-22.
9. Uludağ Ö. Effect of sugammadex and rocuronium combination on cranial neurotoxicity in rats: Experimental study. KafkasUniv Vet Fak Derg 2019; 25: 793-9.
10. Miller MA, Zachary JF. Mechanisms and morphology of cellular injury, adaptation, and death. In: Pathologic Basis of Veterinary Disease. Elsevier; 2017. p. 2-43.e19.
11. Uchiyama M, Mihara M. Determination of malonaldehyde precursor in tissue by TBA test. AnalBiochem1978; 86: 271-8.
12. Elman GL. Tissue sulphydryl groups. Arch Biochem Biophys 1959; 82: 70-7.
13. Cammu G, De Kam PJ, Demeyer I, et al. Safety and tolerability of single intravenous doses of sugammadex administered simultaneously with rocuronium or vecuronium in healthy volunteers. Br J Anaest 2008; 100: 373–9.
14. De Boer HD, Driessen JJ, Marcus MA, Kerkkamp H, Heeringa M, Klimek M. Reversal of rocuronium-induced (1.2 mg/kg) profound neuromuscular block by sugammadex: A multi center, dose-finding and safety study. Anesthesiology 2007; 107: 239–44.
15. Osaka Y, Shimada N, Satou M, et al. A case of atrioventricular block (Wenckebach type) induced by sugammadex. J Anesth 2012; 26: 627–8.
16. Kokki M, Ali M, Turunen M, Kokki H. Suspected unexpected adverse effect of sugammadex: Hypotension. Eur J Clin Pharmacol 2012; 68: 899–900.
17. Bhavani SS. Severe bradycardia and asystole after sugammadex. Br J Anesth 2018; 121: 95–6.
18. Tsur A, Kalansky A. Hypersensitivity associated with sugammadex administration: a systematic review. Anaesthesia 2014; 69: 1251–7.
19. Dahl V, Pendeville PE, Hollmann MW, Heier T, Abels EA, Blobner M. Safety and efficacy of sugammadex for the reversal of rocuronium-induced neuromuscular blockade in cardiac patients undergoing noncardiac surgery. Eur J Anaesthesiol 2009; 26: 874-84.
20. de Kam PJ, van Kuijk J, Prohn M, Thomsen T, Peeters P. Effects of sugammadex doses up to 32 mg/kg alone or in combination with rocuronium or vecuronium on QTc prolongation: a thorough QTc study. Clin Drug Investig 2010; 30: 599–611.
21. Bostan H, Kalkan Y, Tomak Y, et al. Reversal of rocuronium-induced neuromuscular block with sugammadex and resulting histopathological effects in rat kidneys. Ren Fail 2011; 33: 1019-24.
22. Kalkan Y, Bostan H, Tumkaya L, et al.The effect of rocuronium, sugammadex, and their combination on cardiac muscle and diaphragmatic skeletal muscle cells. J Anesth 2012; 26: 870-7.
23. Tercan M, Yılmaz İnal F, Seneldir H, Kocoglu H. Nephroprotective efficacy of sugammadex in ischemia-reperfusion injury: an experimental study in a rat model. Cureus. 2021; 13: e15726.
24. Ozbilgin S, Yılmaz O, Ergur BU, et al. Effectiveness of sugammadex for cerebral ischemia/reperfusion injury. Kaohsiung J Med Sci 2016; 32: 292-301.

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Overview of peritoneal dialysis outcomes in Northern Cyprus: a nation based study

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ABSTRACT

Aim: Peritoneal dialysis (PD) is the more preferred renal replacement therapy (RRT) option for most patients with end-stage renal disease (ESRD) who can not achieve a kidney transplantation. By an enhanced national PD program, more patients may have a chance to take the advantage of this treatment option. In this study, we aim to investigate whether PD is preferred as the first dialysis treatment modality in Northern Cyprus and reveal the outcomes in order to improve the further PD program.

Material and Method: Peritoneal dialysis patients aged above 18 years who were followed between 2003-2021 in Northern Cyprus were investigated retrospectively. Demographic data, primary kidney diseases, comorbidities and previous RRT modalities were analysed. Episode(s) of peritonitis, total duration on PD therapy, switch to kidney transplantation or HD, and mortality rate were evaluated. Outcomes of elderly (≥ 65 years) and diabetic patients who were dropped out from PD were found out.

Results: A total of 123 patients aged 18-83 years were included in the study. Forty percent of 123 PD patients initiated RRT with PD. The median time was found 1 month in the rest of the patients to transfer PD from other modalities. Renal transplantation was performed in 16.8% of patients during follow-up whereas 34.7% were transferred to HD after 41 months. Inadequate dialysis (40.7%), PD-related infections (29.6%), ultrafiltration insufficiency (18.5%), mechanical complications (11.1%) were the most common causes of switching from PD to HD. The duration of being under PD therapy was 36.5 months for 46 patients who died under PD therapy. Cardiovascular events (50%) and various infections (%35) were the leading causes of mortality. A total of 48 (43.2%) PD patients had at least one episode of peritonitis. Total duration on PD treatment, PD rate as initial RRT modality, the prevalence of hypertension and diabetes mellitus (DM), peritonitis rate, and use of automated PD were not significantly different between elderly (n=34) and adult individuals (18-65 years). The duration of being under PD therapy was longer for males than females (p=0.044) and the duration of PD therapy was similar in the dropped out patients with or without DM (p=0.205).

Conclusion: A significant amount of patients received HD before initiation of PD (60%). Age is not be considered as a challenging barrier for PD initiation in Northern Cyprus. Precautions to prevent the development of peritonitis may contribute to extend the total duration of PD treatment.

Keywords: End-stage renal disease, peritoneal dialysis, renal replacement therapy

The study was presented as an oral presentation in 23rd National Hypertension and Kidney Diseases Congress (2021) and accepted as a mini-oral presentation in 59th European Renal Association (ERA) Congress (2022).

INTRODUCTION

The prevalence of patients with end-stage renal disease (ESRD) undergoing renal replacement therapies (RRTs) is increasing worldwide (1). Although transplantation is the best option among RRTs, there is a shortage of organ donors, and thus most of the patients with ESRD have to be put on either hemodialysis (HD) or peritoneal dialysis (PD) while on a waiting list for transplantation (2). Epidemiology of ESRD and treatment options for these patients should be considered in the health system projections of each country.

Patients should be well informed about different treatment modalities in the predialysis period. Besides the medical factors, patients' preferences, expectancy, and needs should be considered for a better quality of life (3). Modality conversion may sometimes be required during the follow-up. Although the proportion of patients on HD is much more than patients on PD, the pattern of choice may be different among countries and regions (4).

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Analysis of the PD registry of each country may help to improve the national PD program –to determine medical expenses, and demand for health care professionals, to allocate health care resources fairly. With the national PD program, more patients may have a chance to take advantage of this treatment option. Mortality and morbidity data may allow us to take necessary precautions. This may give us a chance to overcome barriers and build a strong PD program to improve health outcomes. In order to contribute to the development of the PD program in Northern Cyprus, we planned to assess our national PD registry as a first step. In this study, we aim to investigate whether PD is preferred as the first dialysis treatment modality in Northern Cyprus and to evaluate the duration of being under PD therapy, and outcomes in order to improve the PD program.

MATERIAL AND METHOD

The study was approved by Dr. Burhan Nalbantoğlu State Hospital Ethics Committee (Date: 5.11.2021, Decision number: 55/21). All procedures were performed in accordance with the principles stated in the Declaration of Helsinki. We evaluated all 125 adult PD patients (> 18 years) who were followed in Northern Cyprus from the year 2003 to 2021. Two missing PD patients were excluded due to moving to another country within 3 months following the onset of the PD therapy. There was a single team who was responsible for follow-up PD patients in the country.

Demographic data, primary kidney diseases, and comorbidities including diabetes mellitus (DM), hypertension, heart failure, peripheral vascular disease, cerebrovascular disease, liver disease, peptic ulcer, malignancy, acquired immunodeficiency syndrome, a solid tumor, or hematological malignancy were noted. Charlson's comorbidity index was calculated for each participant. Previous RRT histories of the patients were screened retrospectively to determine whether PD was the initial RRT modality or not. Duration of time spent on other RRT modalities before initiation of PD was found out. Data of initial peritoneal equilibrium test results and switch to automated PD were collected. The proportion of patients who had peritonitis was determined.

The primary endpoint of this study was PD dropout. PD dropout was defined as the termination of PD therapy and resulted in switching to another RRT (renal transplantation or HD) or mortality. Causes of switching from PD to HD were analyzed. We evaluated patient survival and described the causes of mortality. Peritoneal dialysis duration was calculated for each participant who dropped out from PD. The dropout rates of 1, 2, and 5 years following the onset of the PD initiation were found out. Peritoneal dialysis duration and a dropout rate of 1 and after 5 years of PD initiation were compared between female and male PD patients.

Elderly patients who initiated PD at age ≥65 were compared with adult individuals in terms of initial RRT modality, time passed to initiate PD after first RRT, use of automated PD. Peritoneal dialysis duration, peritonitis rate, use of automated PD, the prevalence of hypertension, diabetes, Charlson's comorbidity index, and outcomes were evaluated for elderly and adult patients who were dropped out from PD. The dropout rates of 1 and 5 years were evaluated for elderly and adult individuals.

Initial RRT modalities, the duration of being under PD therapy, peritonitis rate, use of APD, the prevalence of hypertension, Charlson comorbidity index, and a dropout rate of 1 and 5 years were compared between PD patients with and without DM.

Statistical Analysis

We used the Statistical Program for Social Science (SPSS) version 15 for Windows. Normality tests were performed to identify whether variables were in the normal distribution. Descriptive statistics are presented as mean±standart deviation, median, minimum and maximum values. Categorical values are described as percentages. Student's t-test or Mann-Whitney U test was used for the mean comparison of two groups. Chi-square statistics were used to compare two categorical variables. Binary logistic regression was performed to evaluate the impact of peritonitis on the drop out rate of PD. p value lower than 0.05 was considered statistically significant.

RESULTS

A total of 123 patients aged 18-83 years were included in the study. Twenty-five patients were actively on PD therapy, 2 moved to another country on PD therapy, and 96 dropped out from PD. A greater number of male than female patients received PD (73% vs 27%). Unknown etiology (32.5%) and DM (29.3%) were the most common etiological factors for renal diseases in these patients (**Table**). Peritoneal dialysis was the initial RRT modality for 40% of patients (**Figure 1**). The median time was found 1 month for the patients to transfer PD from other modalities. Automated PD was used by 58 (53.2%) patients.

Table. Demographic and clinical features of peritoneal dialysis patients	
Gender, female/male, n (%)	33/90 (27/73)
Age initiated peritoneal dialysis*	59 (18-83)
Time passed to initiate peritoneal dialysis after first renal replacement therapy, months*	1 (0-328)
Diabetes mellitus prevalence, n (%)	53 (43.4)
Hypertention prevalence, n (%)	104 (87.4)
Primary kidney disease, n (%)	
Diabetes mellitus	36 (29.3)
Glomerulonephritides	17 (13.8)
Hypertension	7 (5.7)
Tubulointerstitial disease	4 (3.3)
Policystic kidney disease	8 (6.5)
Other	10 (8.1)
Unknown	40 (32.5)
Charlson comorbidity index*	5 (2-9)
Initial peritoneal equilibrium test category, n (%)	
Low transporter	3 (3.5)
Low-average transporter	23 (27.1)
High-average transporter	42 (49.4)
High transporter	17 (20)
Patients experienced peritonitis, n (%)	48 (43.2)
Use of automated peritoneal dialysis, n (%)	58 (53.2)
*median (minimum-maximum)	

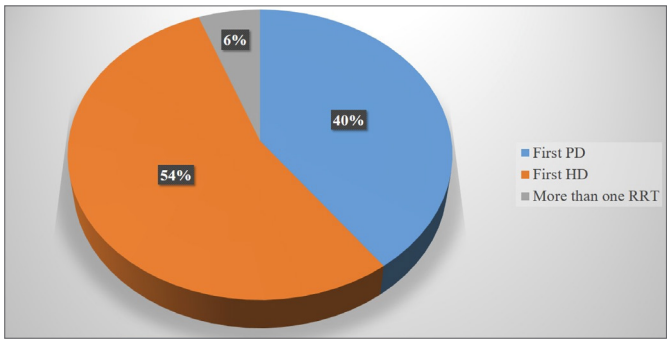

Figure 1. Distrubition of patients according to first renal replacement therapies
*PD; peritoneal dialysis, HD; hemodialysis, RRT; renal replacement therapy

Sixteen (16.8%) patients had renal transplantation during follow-up. Thirty-three patients (34.7%) were switched to HD. Median transfer time to HD was 41 months (1-122 months). Dropout rate to HD was 9.5% in 2019, 4.2% in 2020 and 4.0% in 2021. Inadequacy dialysis and PD-related infections were the most common causes of switching from PD to HD (**Figure 2**). A total of 48 (43.2%) PD patients had at least one episode of peritonitis. Peritonitis was found as an important factor affecting the dropout rate of PD (p<0.0001).

Forty-six (48.4%) patients died on PD therapy at the age of 67±11. Average time spent under PD was found 36.5 months (1-150 months) for these patients. Mortality rates for last three years were evaluated and it was 13.6% in 2019, 14.8% in 2020, and 17.2% in 2021. Cardiovascular events (50%) and various infections (%35) were the leading causes of mortality.

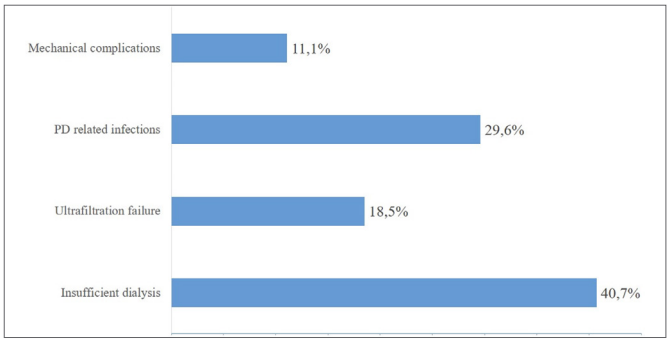

Figure 2. Causes of transfer from peritoneal dialysis to hemodialysis
*PD; peritoneal dialysis

The duration of being under PD therapy was 36 (1-150) months in patients who dropped out from PD. 18.9% (18/95) of dropouts occurred within 1 year of PD initiation, whereas 66.3% (63/95) occured after 2 years and 28.4% (27/95) after 5 years of PD initiation.

Gender

Total duration on PD treatment was longer in males (n=67) then females (n=28) (41 [1-150] vs 21 [3-120] months, p=0.044). Thirty-two percent (9/28) of female patients dropped out within the first year of PD whereas only 13% (9/67) of male patients dropped out of PD within the same period (p=0.034). The dropout number of female and male patients after 5 years of the onset of the PD was similar (6/28 F, 21/67 M, p>0.05).

Elderly and Adult Patients

Elderly patients, who initated PD at aged ≥65, consisted 32.5% (40/123) of all PD patients. Initial RRT modality was PD in 32.5% (13/40) of elderly patients and 43.4% (36/83) of adult patients (p>0.05). Time passed to initiate PD after first RRT was similar between elderly (n=40) and adult patients (n=83) (1.5 [0-328] vs 1[0-164] months, p>0.05). Elderly patients used automated PD more commonly than adult individuals (67% [24/36] vs 47% [34/73], p=0.048).

The duration of being under PD therapy was 34.5 (1-150) months for elderly (n=34) and 38 (1-139) months for adult individuals (n=61) who were dropped out from PD (p>0.05). Peritonitis was similar in elderly and adult patients who were dropped out from PD (16/31 vs 27/54, p>0.05). The use of automated PD was not significantly different between elderly and adult patients who were dropped out from PD (19/30 vs 28/53, p>0.05 for all). Prevalence of hypertension (27/33 vs 56/60, p>0.05) and DM (19/34 vs 26/62, p>0.05) were similar between the two groups. The comorbidity index was found significantly higher in the elderly group than adult individuals who were dropped out (5.85±1.48 vs 4±1.59, p<0.0001). The dropout rate within 1 year of PD was similar for elderly and adult individuals (8/34 vs 10/61, p>0.05). The dropout rate of PD at 5 years was similar for elderly and adult individuals (8/34 vs 19/61, p>0.05).

Diabetic and Non-diabetic Patients

A total of 53 PD patients out of 123 (43.4%) had DM. Diabetic patients who chose PD as the initial RRT modality were not significantly different than non-diabetic patients (n=24/53 vs 25/69, p>0.05).

Patients who dropped out from PD were grouped as diabetic (45/96, 46.9%) and non-diabetic patients (51/96, 53.1%). Peritoneal dialysis duration was 31 (1-139) months for patients with DM and 43 (2-150) months for patients without DM (p>0.05). The prevalence of peritonitis was similar in the two groups (21/39 vs 22/46, p>0.05). The use of APD was similar for patients with and without DM (18/36 vs 29/47, p>0.05). Hypertension was more prevalent in patients with DM than patients without DM (43/44 vs 40/49, p=0.012). The comorbidity index was significantly higher in patients with DM than patients without DM (5.93±1.34 vs 3.48±1.25, p<0.0001). The dropout rate within 1 year of PD onset was similar for diabetic and nondiabetic individuals (7/45 vs 11/50, p>0.05). The dropout rate after 5 years of PD initiation was similar for individuals with and without DM (9/45 vs 18/50, p>0.05).

DISCUSSION

Our present results show that a significant amount of patients required HD before initiation of PD in Northern Cyprus. This was higher than the rates reported in previous studies from other countries (5). Late admission of patients to the nephrology department, delay in predialysis education, less use of urgent PD may be some of the possible explanations for this observation. In this study, we found that patients aged ≥ 65 years had similar results to adults, and diabetic patients had similar outcomes to non-diabetic patients. Therefore, age and DM should not be considered as challenging barriers for PD.

PD has the advantage of being home-based therapy away from the hospital, with social distancing without need for transportation which becomes more important nowadays due to the COVID-19 pandemic (6). Peritoneal dialysis offers patients the opportunity to dialysis with relative protection of residual kidney function and no need for vascular access site. Clinical outcomes and survival were as good as HD (7-9). A more flexible schedule of PD may provide greater patient freedom like more opportunities to travel, and social rehabilitation, and may deliver improved quality of life (4,8,10,11). Reduced early graft dysfunction following transplantation was reported in PD patients (12). Besides all these potential benefits, PD is also accepted as a cost-effective RRT method (13).

Despite all its advantages, the use of PD is lower than HD in most parts of the world (4,7,11,14). Similarly, PD is less likely preferred RRT modality in Northern Cyprus. We need to clarify the reasons why PD is not widely used. There may be factors related to patient, health care team, and health care system (4). Patients may be inadequately educated on modality choice and they may feel fear that leads to reluctance in accepting PD (15). Problems faced arranging catheter insertion and catheter dysfunction as an early technique failure are important challenges (7). Patient education and training may be time-consuming, and a lack of motivation of the health care team may be some of the underlying factors (15). Reimbursement schemes and government policies may be also responsible for the wide variation in the use of PD between countries (4).

Patients with ESRD should have the opportunity to access various treatment options (14). Predialysis patients should be educated timely about the dialysis modalities and encouraged to make a choice that is most suited to their lifestyles (4,7,16). Enough time should be spent discussing treatment modalities and effective educational programs may include sessions with family members (16). Catheter placement, patient training, and continued support may be some of the other factors on which we need to focus (7). A dedicated staff who will participate in the education and training PD programs is essential. The health care team should help patients to feel confident to dialyze. Appropriate socio-economic circumstances and social support are also required, and home visits may support patient cooperation (7).

Patients may admit in late-stage requiring dialysis. Unplanned RRT initiation may lead patients to choose HD instead of PD. These patients are less likely to change their modality to PD. The feeling of improvement that patients feel after starting HD may lead to the perception that the current treatment is the best treatment. An urgent start PD program is an acceptable option and may reduce central venous catheter requirement for patients who prefer to be on PD (7,9). Incidences of catheter-related bloodstream infections and dialysis-related complications were reported lower in urgent-start PD than urgent-start HD via a central venous catheter (3). Patients who have already initiated HD therapy should also receive information about other RRT modalities. A significant number of these patients may transfer to PD in the first 6 months of therapy (16,17). We found that PD was the initial RRT modality only for 40% of patients, and it took approximately 1 month for others to initiate PD after the first RRT. Pulliam et al. reported 25%– 27% of PD patients were transferred from HD, and only 1%–2% had a failed allograft (18). This indicates that PD should be more encouraged as a first RRT modality in

our country. High prevalence of unknown etiology in our cohort may implicate late admission of the patients which may limit time for adequate education in the predialysis period. Urgent PD may also be less performed.

Dropout rate from PD to HD improved in last three years from 9.5% in 2019 to 4.0% in 2021 in Northern Cyprus. Strategies to improve PD technique survival may help to keep patients on PD and lead to higher-quality dialysis with better outcomes (4,7). High rates of technique failure may limit time spent on PD. Unfortunately, the majority of the patients discontinue PD within 2-3 years of commencement (7,11), and only a few patients are left on PD after 5 years of PD initiation (10,18). The longevity of PD still remains a concern (4). Catheter dysfunction, peritoneal or catheter-related infections, inadequacy dialysis, or ultrafiltration failure are among the common reasons for technique failure. Psychosocial barriers like a burden and patient preference are the other contributing factors (19,20). In our study majority of PD patients dropped out after 2 years of PD initiation. Peritoneal dialysis duration was 41 months for patients who dropped out from PD to HD, and insufficient dialysis and PD-related infections were the most common causes. We may need to give greater attention to prevent technique failure to make long-term PD possible. Mortality rate of PD patients was increased in last three years from 13.6% in 2019 to 17.2% in 2021 which might be attributed to COVID-19 pandemia. Cardiovascular events and infections are the leading causes of mortality in PD patients.

The predominance of male patients was noted in our study. There are conflicting reports about gender-specific differences in technique failure. Some trials reported lower technical failure rates in females compared to men (21,22) whereas no gender-specific differences were found in others (23,24). Different characteristics of the PD population like age, the need for sufficient numbers of caregivers might affect the results. Male and female patients may also have different coping skills (25). We found the duration of being under PD therapy was longer in males than females. The dropout rate of female patients was higher within the first year of PD onset, whereas the dropout rate after 5 years of PD onset was similar in the two groups.

The elderly are less likely to initiate RRT with PD than HD (26,27). However, PD may be a better option for the elderly rather than HD as hemodynamic instability and vascular access may be greater problems for this age group. Initiating RRT with PD rather than HD was found associated with better patient survival for the full 5 year follow-up period for patients aged above or below 65 years (4). Availability of assisted PD programs may be useful for some elderly patients who prefer to perform PD but need help (28). Our results supported that elderly patients have comparable outcomes to adult individuals. Elderly patients used automated PD more commonly than adult individuals in our study.

Diabetes is the most common cause of ESRD worldwide. Diabetic patients were less likely to receive PD as first RRT than nondiabetic patients in Europe (26,27,29). The presence of diabetic retinopathy and peripheral neuropathy, anxiety about poorer glycemic control, and increased risk for peritonitis may be the underlying reasons (29). Transfer to HD may be more common in diabetic PD patients (25). However, our study revealed that a significant amount of PD patients in Northern Cyprus had DM, and number of diabetic patients who chose PD as the initial RRT modality were not different than non-diabetic patients. Despite the greater prevalence of comorbidities, the duration of being under PD therapy and dropout rate were similar in patients with and without DM. Peritonitis rate was not different as well. All these results encourage the use of PD in diabetic patients. The difference in results may be due to the heterogeneity of patients with DM.

The limitations of the study are as follow. First, psychosocial factors could not be evaluated among the dropout causes due to the retrospective design of the study. Second, data about predialysis education, nephrology care before the start of dialysis, and the presence of caregivers were not included. Third, as this is a nation-based study, results may not be entirely generalized.

CONCLUSION

A significant amount of patients required HD before initiation of PD. Early admission of patients to the nephrology department and timely initiation of education programs may contribute to decreasing the need for HD before PD. As elderly patients had similar results with adult individuals, age is not be considered as a challenging barrier for PD. Precautions to prevent the development of peritonitis may contribute to extend the total duration of PD treatment. Region-specific issues related to using PD need to be described to improve the outcomes of PD patients.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was approved by Dr. Burhan Nalbantoğlu State Hospital Ethics Committee (Date: 5.11.2021, Decision number: 55/21).

Informed Consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

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Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

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REFERENCES

1. GBD Chronic Kidney Disease Collaboration. Global, regional, and national burden of chronic kidney disease, 1990-2017: a systematic analysis for the Global Burden of Disease Study 2017. Lancet 2020; 395: 709-33.
2. Tang SCW, Lai KN. Peritoneal dialysis: the ideal bridge from conservative therapy to kidney transplant. J Nephrol 2020; 33: 1189-94.
3. Van Biesen W, Lameire N. Increasing peritoneal dialysis initiation worldwide: 'there are none so blind as those who will not see'. Nephrol Dial Transplant 2020; 35: 1458-61.
4. Li PK, Chow KM, Van de Luijtgaarden MW, et al. Changes in the worldwide epidemiology of peritoneal dialysis. Nat Rev Nephrol 2017; 13: 90-103.
5. Pulliam J, Li NC, Maddux F, Hakim R, Finkelstein FO, Lacson E Jr. First-year outcomes of incident peritoneal dialysis patients in the United States. Am J Kidney Dis 2014; 64: 761-9.
6. Ikizler TA, Kliger AS. Minimizing the risk of COVID-19 among patients on dialysis. Nat Rev Nephrol 2020; 16: 311-3.
7. Briggs V, Davies S, Wilkie M. International Variations in Peritoneal Dialysis Utilization and Implications for Practice. Am J Kidney Dis 2019; 74: 101-10.
8. Subramanian S, Javaid MM. Peritoneal dialysis as the dialysis initial modality of choice for renal replacement therapy initiation: moving from "Why" to "Why Not". Blood Purif 2017; 44: 106-7.
9. Mendes ML, Alves CA, Bucuvic EM, Dias DB, Ponce D. Peritoneal dialysis as the first dialysis treatment option initially unplanned. J Bras Nefrol 2017; 39: 441-6.
10. Chaudhary K, Sangha H, Khanna R. Peritoneal dialysis first: rationale. Clin J Am Soc Nephrol 2011; 6: 447-56.
11. Ito Y, Tawada M, Tine S, Mizuno M, Suzuki Y, Katsuno T. Current status of peritoneal dialysis in Japan. Contrib Nephrol 2018; 196: 123-8.
12. Joachim E, Gardezi AI, Chan MR, Shin JI, Astor BC, Waheed S. Association of pre-transplant dialysis modality and post-transplant outcomes: a meta-analysis. Perit Dial Int 2017; 37: 259-65.
13. Treharne C, Liu FX, Arici M, Crowe L, Farooqui U. Peritoneal dialysis and in-centre haemodialysis: a cost-utility analysis from a UK payer perspective. Appl Health Econ Health Policy 2014; 12: 409-20.
14. Stel VS, de Jong RW, Kramer A, et al. Supplemented ERA-EDTA Registry data evaluated the frequency of dialysis, kidney transplantation, and comprehensive conservative management for patients with kidney failure in Europe. Kidney Int 2021; 100: 182-95.
15. van Biesen W, Veys N, Lameire N, Vanholder R. Why less success of the peritoneal dialysis programmes in Europe? Nephrol Dial Transplant 2008; 23: 1478-81.
16. Devoe DJ, Wong B, James MT, et al. Patient education and peritoneal dialysis modality selection: a systematic review and meta-analysis. Am J Kidney Dis 2016; 68: 422-33.
17. Blake PG, Quinn RR, Oliver MJ. Peritoneal dialysis and the process of modality selection. Perit Dial Int 2013; 33: 233-41.
18. Guo A, Mujais S. Patient and technique survival on peritoneal dialysis in the United States: evaluation in large incident cohorts. Kidney Int Suppl 2003; 3-12.
19. See EJ, Johnson DW, Hawley CM, et al. Risk predictors and causes of technique failure within the first year of peritoneal dialysis: an Australia and New Zealand Dialysis and Transplant Registry (ANZDATA) Study. Am J Kidney Dis 2018; 72: 188-97.
20. Navaratnarajah A, Clemenger M, McGrory J, et al. Flexibility in peritoneal dialysis prescription: Impact on technique survival. Perit Dial Int 2021; 41: 49-56.
21. Shen JI, Mitani AA, Saxena AB, Goldstein BA, Winkelmayer WC. Determinants of peritoneal dialysis technique failure in incident US patients. Perit Dial Int 2013; 33: 155-66
22. Kitterer D, Segerer S, Braun N, Alscher MD, Latus J. Gender-Specific Differences in Peritoneal Dialysis. Kidney Blood Press Res 2017; 42: 276-83.
23. Chidambaram M, Bargman JM, Quinn RR, Austin PC, Hux JE, Laupacis A. Patient and physician predictors of peritoneal dialysis technique failure: a population based, retrospective cohort study. Perit Dial Int 2011; 31: 565-73.
24. Mujais S, Story K. Peritoneal dialysis in the US: evaluation of outcomes in contemporary cohorts. Kidney Int Suppl 2006; : S21-6.
25. Zhang L, Lee WC, Wu CH, et al. Importance of non-medical reasons for dropout in patients on peritoneal dialysis. Clin Exp Nephrol 2020; 24: 1050-7.
26. van de Luijtgaarden MW, Jager KJ, Segelmark M, et al. Trends in dialysis modality choice and related patient survival in the ERA-EDTA Registry over a 20-year period. Nephrol Dial Transplant 2016; 31: 120-8.
27. van de Luijtgaarden MW, Noordzij M, Stel VS, et al. Effects of comorbid and demographic factors on dialysis modality choice and related patient survival in Europe. Nephrol Dial Transplant 2011; 26: 2940-7.
28. Boyer A, Solis-Trapala I, Tabinor M, Davies SJ, Lambie M. Impact of the implementation of an assisted peritoneal dialysis service on peritoneal dialysis initiation. Nephrol Dial Transplant 2020; 35: 1595-601.
29. Maruyama Y, Higuchi C, Io H, et al. Comparison of peritoneal dialysis and hemodialysis as first renal replacement therapy in patients with end-stage renal disease and diabetes: a systematic review. Ren Replace Ther 2019; 5: 44.

Original Article

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The effect of empagliflozin on monocyte high-density lipoprotein ratio in patients with type 2 diabetes mellitus

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ABSTRACT

Aim: We aimed to investigate the effect of empagliflozin, which is started in patients with type 2 diabetes mellitus (T2DM), on neutrophil leumphocyte ratio (NLR) and monocyte high-density lipoprotein ratio (MHR), which are used as inflammation, glycemic control and oxidative markers.

Material and Method: The file systems of T2DM patients who used empagliflozin for at least 12 weeks were retrospectively analyzed. Demographic data of the patients were recorded. biochemical and hemogram parameters were compared before and after empagliflozin.

Results: 194 patients were included in the study. Plasma fasting glucose ($p<0.001$), hemoglobin A1c (HbA1c)($p<0.001$), low-density lipoprotein cholesterol (LDL-C) ($p=0.041$), NLR($p=0.002$) and MHR($p=0.042$) values of T2DM patients after empagliflozin treatment were statistically significantly decreased compared to pre-treatment with empagliflozin. HDL-C value($p=0.003$), on the other hand, increased significantly after empagliflozin

Conclusion: NLR and MHR are inexpensive and practical markers of inflammation. This result shows us that NLR and MHR should be used as inflammation markers in patients using empagliflozin.

Keywords: Empagliflozin, MHR, NLR

INTRODUCTION

Type 2 diabetes mellitus (T2DM) is a progressive disease with a complex pathophysiology. Increasing insulin resistance, progressive deterioration of β-cell function, dysfunctional adipocytes, gastrointestinal incretin defects, increased glucose reabsorption from the kidneys, hyperglucagonemia, and neurotransmitter dysfunction may contribute to development of diabetesGlucose control is a central focus in the management of T2DM, and reducing hyperglycemia has been shown to decrease microvascular complications of diabetes (1). Chronic inflammation plays an important role in the pathogenesis of diabetes, its development and complications (2,3). Studies show that the levels of inflammatory cytokines such as CRP, IL-1, IL-6, TNF-α increase in patients with diabetes (4). Neutrophil/lymphocyte ratio (NLR) is an indicator of systemic inflammation and is accepted as a marker of inflammation in complications such as microvascular and macrovascular in diabetic patients (5). The monocyte-to-high density lipoprotein ratio (MHR) has recently been implemented as an indicator of inflammation and oxidative stress. MHR indicates inflammation and oxidative stress due to the proinflammatory effect of the monocytes, as well as the anti-inflammatory and antioxidant effect of the high-density lipoprotein cholesterol (HDL-C). Several studies have used these metrics to determine whether inflammation and atherosclerosis contribute to the etiopathogenesis of cardiovascular and cerebrovascular diseases. MHR has been found to be significant as a biomarker in the development of microvascular complications in diabetic patients (6,7). Empagliflozin, a sodium-glucose cotransporter 2 (SGLT2) inhibitor, is a newly developed oral antidiabetic drug to enhance renal glucose excretion or glycosuria and reduce hyperglycaemia in an insulin-independent manner by highly selective inhibition of SGLT2. SGLT2 is mainly located in the apical brush border membrane of the S1 segment of the proximal convoluted tubules, which regulates 90% of the reabsorption of glucose from glomerular filtrate. Therefore, SGLT2 inhibitor can increase the urinary glucose level and reduce blood glucose. Empagliflozin is different form conventional antidiabetic drugs, which

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rely on insulin secretion, and represents a novel class of antidiabetic drugs. It has been approved for the treatment of type 2 diabetes in adults since 2014 (8,9). In our study, we aimed to investigate the effect of empagliflozin, which is started in patients with Type 2DM, on NLR and MHR, which are used as inflammation, glycemic control and oxidative markers.

MATERIAL AND METHOD

This study was planned retrospectively. The study was carried out with the permission of Hitit University Medical Faculty Non-interventional Clinical Researches Ethics Committee (Date: 29.12.2021, Decision No: 2021-88). All procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki.

Patients who applied to our internal medicine outpatient clinic between 01.01.2019 and 01.01.2022 with a diagnosis of T2DM, who were started on empagliflozin 10 mg and used for at least 12 weeks were screened. 194 patients who met the inclusion criteria were included in the study. Inclusion criteria for the study; Patients who have not used empagliflozin before and have received empagliflozin treatment for at least 12 weeks, patients over the age of 18, patients under 75 years of age, patients who are not pregnant, those without acute infection, those who have not started antihyperlipidemia treatment in the last month and have not changed, and those who do not use drugs affecting the bone marrow, spleen-related disease, acute and chronic inflammatory disease and consists of people without a history of malignancy. Exclusion criteria; Type-1 DM, who use empagliflozin for less than 12 weeks, pregnant women and patients with chronic renal failure. Demographic data, comorbidities and diabetes medication used before empagliflozin were noted. Glucose, hemoglobin A1c (HbA1c), HDL-C, low-density lipoprotein cholesterol (LDL-C), total cholesterol, triglyceride, alanine aminotransferase (ALT), aspartate aminotransferase (AST) and hemogram parameters were recorded before and after empagliflozin.

This study was approved by the university/local human research ethics committee, and all procedures performed in studies involving human participants were conducted in accordance with the ethical standards of the institutional and/or national research committee, the 1964 Declaration of Helsinki and subsequent amendments or comparable ethical standards.

Statistical Analysis

IBM SPSS 22 program was used for data analysis and statistical analysis. Student's T test was used for intergroup comparisons of normally distributed parameters as well as descriptive statistical methods (mean, standard deviation, ratio) when evaluating study data; Mann-Whitney U test was used for the intergroup comparisons of non-normally distributed parameters. In addition, Wilcoxon Signed Rank test was used. The results were evaluated at the 95% confidence interval and the significance level of $p<0.05$.

RESULTS

A total of 194 patients, 86 male and 108 female, using empagliflozin were included in our study. The mean age was 63.12±4.12 in men and 64.26±5.90 in women. The duration of DM was 13.05±5.34 in men and 14.09±6.01 in women. DM drugs used before empagliflozin, comorbidities and complications of DM are shown in **Table 1.**

Table 1. Demographic and clinical data of the patients

Sex=n (%)	
Male	86 (44.3)
Female	108 (55.6)
Age (years)±SD	
Male	63.12±4.12
Female	64.26±5.90
Disease duration (years) ± SD	
Male	13.05±5.34
Female	14.09±6.01
Comorbidity (n)	
Hypertension	105
Hyperlipidemia	47
Coronary artery disease	31
Cerebrovascular disease	7
None	64
DM medication before empagliflozin(n)	
Insulin	75
Metformin	49
Linagliptin	24
Vildagliptin	28
Pioglitazone	27
Other	10
DM microvascular complications(n)	
None	94
Neuropathy	39
Nephropathy	29
Retinopathy	32

Plasma fasting glucose, HbA1c, LDL-C, NLR and MHR values of T2DM patients after empagliflozin treatment were statistically significantly decreased compared to pre-treatment with empagliflozin ($p<0,05$). HDL-C value, on the other hand, increased significantly after empagliflozin ($p<0,05$) (**Table 2**).

There was no statistically significant difference in blood parameters of T2DM patients with neuropathy after using empagliflozin ($p>0,05$) (**Table 3**).

Table 2. Comparison of biochemical and hemogram parameters of T2DM patients before and after empagliflozin

	Before Empagliflozin Mean±SD	After Empagliflozin Mean±SD	p
Plasma fasting glucose (mg/dL)	188.02±63.80	146.69±57.04	<0.001*
HbA1c (%)	8.24±1.83	6.37±1.43	<0.001*
Triglyceride (mg/dl)	208.51±72.28	192.60±60.17	0.063
Total Cholesterol (mg/dl)	173.66±52.73	171.72±49.24	0.071
HDL-C (mg/dl)	41.82±11.83	46.64±13.61	0.003*
LDL-C(mg/dl)	108.91±37.27	101.87±43.28	0.041*
ALT (IU/L)	23.19±11.41	24.18±13.06	0.073
AST (IU/L)	20.93±6.93	21.22±9.54	0.967
White blood cell	8.55±4.64	8.40±3.48	0.816
Hemoglobin (g/dl)	13.08±4.07	13.87±2.27	0.163
Neutrophil(10^3/mm^3)	5.01±1.38	4.93±1.19	0.263
Lymphocyte (10^3/mm^3)	2.62±0.43	2.67±0.58	0.107
Monocyte (10^3/mm^3)	0.49±0.11	0.50±0.73	0.473
NLR	1.92±0.63	1.74±0.78	0.002*
MHR	0.01171±0.00401	0.01072±0.00941	0.042*
Platelet (10^3/mm^3)	298.76±72.34	287.09±71.23	0.118

*p<0.05, SD: standard deviation

Table 3. Comparison of biochemical and hemogram parameters of T2DM patients with diabetic neuropahty before and after empagliflozin

	Before Empagliflozin Mean±SD	After Empagliflozin Mean±SD	p
Plasma fasting glucose (mg/dL)	196.96±54.82	183.69±47.03	0.634
HbA1c (%)	10.04±1.97	9.61±1.23	0.072
Triglyceride (mg/dl)	188.71±62.38	184.59±61.24	0.963
Total Cholesterol (mg/dl)	182.31±46.28	171.81±47.28	0.821
HDL-C (mg/dl)	40.36±10.47	43.75±11.58	0.053
LDL-C (mg/dl)	107.99±34.53	108.78±36.41	0.061
NLR	3.69±0.99	3.36±0.78	0.072
MHR	0.01611±0.00321	0.01539±0.00611	0.143

SD: standard deviation

DISCUSSION

In our study, after empagliflozin treatment in T2DM patients, plasma fasting glucose, HbA1c, LDL-C, NLR and MHR values decreased significantly, while HDL-C values increased significantly. The prevalence of type 2 diabetes mellitus has doubled over the past 3 decades and is likely to affect a half a billion people in the next 3 decades (10). Female gender and advanced age are predisposing factors for T2DM (11). Of the patients participating in our study, 55.6% were female and 44.3% were male. The mean age was found to be 64.01 years.

Sodium glucose cotransporter 2 (SGLT2) inhibitors have a unique mecha-nism of action leading to excretion of glucose in the urine and subsequent lowering of plasma glu-cose. This mechanism is independent of β-cell function; thus, these agents are effective treatment for type 2 diabetes mellitus (T2DM) at theoretically any disease stage. Empagliflozin is one of three approved SGLT2 inhibitors (12). In a study by Rosenstock et al. (13) a statistically significant decrease was found in HbA1c, fasting plasma glucose and body weight after 12 weeks of empagliflozin treatment. In our study, similar to the literature, a statistically significant decrease was found in the fasting plasma glucose and HbA1c averages of the patients after empagliflozin (p<0.001).

SGLT2 inhibitors reduce body weight and visceral adiposity, and improve various metabolic abnormalities associated with metabolic syndrome such as blood pressure, lipid profile, and serum uric acid level (14). SGLT2 inhibitors are associated with a small increase in HDL-C as well as an increase in LDL-C with concomitant reductions in triglyceride levels (15,16). In addition, a meta-analysis of 34 randomized controlled trials showed that the administration of SGLT2 inhibitors increased HDL-C (mean difference 1.93 mg/dL), LDL-C (mean difference 3.5 mg/dL) and decreased serum triglycerides (mean difference 7.8 mg/dL) (17). In the comparison study of empagliflozin 10mg, empagliflozin 25mg and placebo by Hach et al. (18) small increases in HDL-cholesterol and LDL-cholesterol and a small decrease in triglyceride levels were observed in the empagliflozin group. In our results, there was a significant increase in HDL-C and a significant decrease in LDL-C (HDL-C p:0.003, LDL-C p:0.041), while the decrease in triglyceride level was not

statistically significant (p:0.063). In a retrospective study by Yuya et al. (19) in non-alcoholic fatty liver patients, patients using dipeptidyl peptidase-4 inhibitor and SGLT2 inhibitor for 24 weeks were analyzed. Decreases in transaminase activities were found to be similar in both groups. We did not detect any significant changes in AST and ALT values before and after empagliflozin.

MHR and NLR values are used as markers of inflammation in diabetic patients (20,21). There are studies showing the anti-inflammatory and antioxidant properties of empagliflozin treatment in T2DM patients (22,23). However, we could not find any study in the literature investigating the effect of empagliflozin treatment on MHR and NLR levels. Chronic inflammatory disorders and dyslipidemia in type 2 diabetes mellitus (T2DM) are essential contributors to the development of atherosclerotic cardiovascular disease. High NLR and MHR values are associated with cardiovascular disease in T2DM patients (24,25). Cardiovascular mortality is the principal cause of death in individuals with T2DM. The recently published Empagliflozin Cardiovascular Outcome Event Trial in Type 2 Diabetes Mellitus Patients (EMPA-REG OUTCOME) study demonstrated that in T2DM patients with high cardiovascular disease risk empagliflozin reduced the primary major adverse cardiac event end point (cardiovascular death, nonfatal myocardial infarction, nonfatal stroke) by 14% (26). In our study, a significant decrease was found in NLR and MHR values after empagliflozin (NLR p:0.002, MHR p:0.042).

Up to 50% of diabetes patients suffer from microvascular complications, including diabetic peripheral neuropathy, diabetic retinopathy and diabetic kidney disease (27,28). There are also evidences that inflammation may play a key role in occurrence of microvascular complications This result shows us that NLR and MHR should be used as inflammation markers in patients using empagliflozin. (24,29,30). In addition, oxidative stress plays a strong role in the pathogenesis of diabetic complications (31). In the study of the effect of empagliflozin on diabetic microvascular complications, Eid et al. (32) did not find any effect on neuropathy in T2DM subjects. In the study of Mehta et al. (33) patients using empagliflozin did not detect any difference in the improvement of diabetic neuropathy when compared to patients using other oral antihyperglycemic drugs. In our study, we did not find any changes in the biochemical and hemogram parameters of diabetic neuropathy patients using empagliflozin in accordance with the literature.

CONCLUSION

Empagliflozin's decrease in NLR and MHR in type 2 DM patients shows that it provides anti-inflammatory activity in these patients

Limitation

This study has some limitations.

1. It is not known that the patients took antidiabetic drugs regularly before empagliflozin
2. Whether the drug use is regular after empagliflozin treatment
3. Failure to evaluate diabetic neuropathic complaints and findings after empagliflozin

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Hitit University Medical Faculty Non-interventional Clinical Researchs Ethics Committee (Date: 29.12.2021, Decision No: 2021-88).

Informed Consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

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Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper and that they have approved the final version

REFERENCES

1. J Levine M. Empagliflozin for type 2 diabetes mellitus: an overview of phase 3 clinical trials. Curr Diab Rev 2017; 13: 405-23.
2. Xu T, Weng Z, Pei C, et al. The relationship between neutrophil-to-lymphocyte ratio and diabetic peripheral neuropathy in type 2 diabetes mellitus. Medicine (Baltimore) 2017; 96: 8289.
3. Liu S, Zheng H, Zhu X, Mao F, Zhang S, Shi H. Neutrophil-to-lymphocyte ratio is associated with diabetic peripheral neuropathy in type 2 diabetes patients. Diabetes Res Clin Pract 2017; 130: 90-7.
4. De Rooij SR, Nijpels G, Nilsson PM, Nolan JJ, Gabriel R, Bobbioni-Harsch E. Low-grade chronic inflammation in the relationship between insulin sensitivity and cardiovascular disease (RISC) population: associations with insulin resistance and cardiometabolic risk profile. Diabetes Care 2009; 32: 1295-301.
5. Qiao S, Gao W, Guo S. Neutrophil–lymphocyte ratio (NLR) for predicting clinical outcomes in patients with coronary artery disease and type 2 diabetes mellitus: a propensity score matching analysis. Therapeutics and Clinical Risk Management 2020; 16: 437-43.
6. You S, Zhong C, Zheng D, Xu J, Zhang X, Liu H. Monocyte to HDL cholesterol ratio is associated with discharge and 3-month outcome in patients with acute intracerebral hemorrhage. J Neurol Sci 2017; 372: 157-61.
7. Vural G, Gümüsyayla Ş. Monocyte-to-high density lipoprotein ratio is associated with a decreased compound muscle action potential amplitude in patients with diabetic axonal polyneuropathy. Medicine 2018; 97: 12857.

8. Byrne NJ, Parajuli N, Levasseur JL, et al. Empagliflozin prevents worsening of cardiac function in an experimental model of pressure overload-induced heart failure. JACC Basic Transl Sci 2017; 2: 347–54.
9. Li C, Zhang J, Xue M, et al. SGLT2 inhibition with empagliflozin attenuates myocardial oxidative stress and fibrosis in diabetic mice heart. Cardiovascr Diabetol 2019; 18: 1-13.
10. Chen L, Magliano DJ, Zimmet PZ. The worldwide epidemiology of type 2 diabetes mellitus–present and future perspectives. Nat Rev Endocrinol 2011; 8: 228–36.
11. Diyabetes Mellitus ve Komplikasyonlarının Tanı, Tedavi ve İzlem Klavuzu. 2020: Türkiye Endokrinoloji ve Metabolizma Derneği.
12. J Levine M. Empagliflozin for type 2 diabetes mellitus: an overview of phase 3 clinical trials. Curr Diab Rev 2017; 13: 405-23.
13. Rosenstock J, Seman LJ, Jelaska A, et al. Efficacy and safety of empagliflozin, a sodium glucose cotransporter 2 (SGLT2) inhibitor, as add-on to metformin in type 2 diabetes with mild hyperglycaemia. Diabetes, Obesity and Metabolism 2013; 15: 1154-60.
14. Nagahisa T, Saisho Y. cardiorenal protection: Potential of sglt2 inhibitors and glp-1 receptor agonists in the treatment of type 2 diabetes. Diabetes Ther 2019; 10: 1733–52.
15. Monami M, Nardini C, Mannucci E. Efficacy and safety of sodium glucose co-transport-2 inhibitors in type 2 diabetes: a meta-analysis of randomized clinical trials. Diabetes Obes Metab 2014; 16: 457–66.
16. Hardy E, Ptanszynska A, de Bruin TWA. Changes in lipid profiles of patients with type 2 diabetes mellitus on dapagliflozin therapy. Diabetologia 2013; 56: 233.
17. Tentolouris A, Vlachakis P, Tzeravini E, Eleftheriadou I, Tentolouris N. SGLT2 inhibitors: a review of their antidiabetic and cardioprotective effects. Int J Environ Res Public Health 2019; 16: 2965.
18. Hach T, Gerich J, Salsali A, et al. Empagliflozin improves glycemic parameters and cardiovascular risk factors in patients with type 2 diabetes (T2DM): pooled data from four pivotal phase III trials. Diabetologie und Stoffwechsel 2014; 9: 142.
19. Seko Y, Sumida Y, Tanaka S, et al. Effect of sodium glucose cotransporter 2 inhibitor on liver function tests in Japanese patients with nonalcoholic fatty liver disease and type 2 diabetes mellitus. Hepatology Research 2017; 47: 1072-8.
20. Shiny A, Bibin YS, Shanthirani CS, et al. Association of neutrophillymphocyte ratio with glucose intolerance: an indicator of systemic inflammation in patients with type 2 diabetes. Diabetes Technol Ther 2014; 16: 524–30.
21. Onalan E. The relationship between monocyte to high-density lipoprotein cholesterol ratio and diabetic nephropathy. Pakistan J Med Sci 2019; 35: 1081-6.
22. Canet F, Iannantuoni F, Marañon AMD, et al. Does empagliflozin modulate leukocyte–endothelium ınteractions, oxidative stress, and ınflammation in type 2 diabetes? Antioxidants 2021; 10: 1228.
23. Singh RB, Fatima G, Kumar P, et al. Effects of empagliflozin on proinflammatory cytokines and other coronary risk factors in patients with type 2 diabetes mellitus: A single-arm real-world observation. Int J Clin Pharmacol Therapeutics 2021; 59: 17.
24. Wan H, Wang Y, Fang S, et al. Associations between the neutrophil-to-lymphocyte ratio and diabetic complications in adults with diabetes: a cross-sectional study. J Diabetes Res 2020; 2020: 9.
25. Chen JW, Li C, Liu ZH, et al. The role of monocyte to high-density lipoprotein cholesterol ratio in prediction of carotid intima-media thickness in patients with type 2 diabetes. Frontiers in Endocrinology 2019; 10: 191.
26. Zinman B, Wanner C, Lachin JM, et al. Empagliflozin, cardiovascular outcomes, and mortality in type 2 diabetes. New Engl J Med 2015; 373: 2117-28.
27. Feldman EL, Callaghan BC, Pop-Busui R, et al. Diabetic neuropathy. Nat Rev Dis Primers 2019; 5: 41.
28. Winocour PH. Diabetes and chronic kidney disease: An increasingly common multi-morbid disease in need of a paradigm shift in care. Diabet. Med. 2018; 35: 300–5.
29. Öztürk ZA, Kuyumcu ME, Yesil Y, et al. Is there a link between neutrophil-lymphocyte ratio and microvascular complications in geriatric diabetic patients? J Endocrinol Invest 2013; 36: 593-9.
30. DiGangi C. Neutrophil-lymphocyte ratio: Predicting cardiovascular and renal complications in patients with diabetes. Journal of the American Association of Nurse Practitioners 2016; 28: 410-4.
31. Feldman EL, Nave KA, Jensen TS, Bennett DLH. New horizons in diabetic neuropathy: mechanisms, bioenergetics, and pain. Neuron 2017; 93: 1296–313.
32. Eid S A, O'Brien PD, Hinder LM, et al. Differential effects of empagliflozin on microvascular complications in murine models of type 1 and type 2 diabetes. Biology 2020; 9: 347.
33. Mehta S, Nain P, Agrawal BK, et al. Effectiveness of empagliflozin with vitamin D supplementation in peripheral neuropathy in type 2 diabetic patients. Cureus 2021; 13: 12.

Original Article

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Relationship between lipid profile and monocyte to highdensity lipoprotein ratio with disease severity in chronic obstructive pulmonary disease patients

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ABSTRACT

Aim: To examine lipid profile and monocyte to high density lipoprotein ratio ratio (MHR) values in stable chronic obstructive pulmonary disease patients.

Material and Method: Patients admitted to our hospital with the diagnosis of chronic obstructive pulmonary disease (COPD) between 01.01.2014 - 01.01.2020 were included in the study and evaluated retrospectively. According to the COPD guideline, two main groups were formed as A+B and C+D. Demographic characteristics, hemogram, C-Reaktif protein (CRP), albumin, lipid profile values were analyzed.

Result: In our study, there were 360 cases, 293 (81.4%) of which were male. The mean age was 67.61±8.7 years. There were 162 cases (45%) in the A+B group and 198 (55%) in the C+D group.White blood cell (WBC) , neutrophil, lymphocyte, neutrophil/ lymphocyte ratio (NLR), monocytes, hemoglobin, CRP, Albumin, high density lipoprotein (HDL), monocyte to HDL ratio (MHR) were found to be different at the level of statistical significance, while cholesterol, triglyceride and low density lipoprotein (LDL) were not at this level of significance. When evaluated with multivariate regression analysis afterwards, it was observed that the statistical significance levels of MHR, CRP and albumin values continued.

Conclusion: We think that high MHR rate, high CRP, and low albumin values in stable COPD patients may be a stimulant for increased disease severity.

Keywords: Albumin, CRP, COPD, HDL, lipit profile, MHR

INTRODUCTION

COPD is an important chronic disease, the most important cause of which is tobacco use, with significant morbidity and the 3rd most frequent mortality results all over the world. According to the global iniative for chronic obstructive lung disease (GOLD) 2021 guideline, it is divided into A, B, C, D groups from mild to severe clinical according to the Unified COPD assessment determined according to symptoms and exacerbation history (1). The lower the fev1 value, which is the respiratory function parameter, is detected in COPD patients, the higher the mMRC and CAT values, which are symptom indicators. There is a negative correlation between low FEV1 values and group C and D with advanced COPD. This means that FEV1 values are expected to be significantly lower in groups C and D. When evaluated according to the GOLD 2021 guideline, the more advanced the COPD, the higher the morbidity and mortality is expected according to the combined COPD assessment consisting of symptoms and exacerbations (1,2). COPD is a systemic inflammatory disease and may be associated with many diseases, especially cardiovascular diseases (1).

In case of persistence of inflammation in COPD, other organs may also be damaged and related diseases may occur (3,4). The incidence of comorbid metabolic syndrome and low HDL in COPD is higher than in normal healthy individuals (5). There are studies showing that HDL has antiatherogenic, antioxidant and anti-inflammatory properties (6,7). Other lipid abnormalities that tend to accompany low HDL include elevated triglyceride levels, especially in the presence of a high ratio of LDL-C to HDL (6).

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Monocytes are a cell type that increases the inflammatory process and may be associated with increased oxidative stress (8,9). Considering the relationship of both monocytes and HDL with inflammation, there are studies showing that the MHR ratio may be an indicator of systemic inflammation (10,11)

There are limited studies in the literature examining the relationship between COPD lipid profile and there are conflicting results in the relationship between monocyte/HDL ratio, HDL and other lipids, airway restriction and disease severity in COPD (11-16).

For this purpose, our primary aim in our study is to separate the COPD groups as A+B and C+D groups and to analyze the lipid profile and MHR data in terms of their relationship with disease severity and their ability to be a biomarker. Additionally, to examine demographic data, clinical characteristics and other laboratory data.

MATERIAL AND METHOD

Patients who applied to our hospital with the diagnosis of COPD between 01.01.2014-01.01.2020 were included in the study. This study was approved by the Health Sciences University Keçiören Education and Training Hospital Clinical Studies Ethics Board (Date: 22.03.2022, Decision No: 2012-KAEK-15/2490). All procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki. The study group consisted of the patients who met the lipid panel and inclusion criteria as soon as the patients were evaluated in their stable periods. Patients were defined according to the combined COPD assessment classification according to the 2022 GOLD guideline; Two main groups were formed as A+B and C+D groups. Demographic and clinical characteristics and laboratory characteristics were analyzed. In addition, in the context of the combined COPD assessment; Risk factors independent of disease stages were investigated.

Inclusion Criterias

COPD over 40 years of age, having a smoking history of at least 10 years or more; COPD stability for at least 1 month ; having no acute infection; basic demographic features, pulmonary function test (PFT), body mass index (BMI), hemogram, CRP, albumin data; cholesterol, triglyceride, HDL, LDL, hemogram, CRP, albumin values taken in the outpatient follow-up visit; not taking lipid-lowering medications; no known atherosclerotic heart diseases; non-diabetic; without malignancy; additional cases without lung diseases, were included in the study (**Figure**).

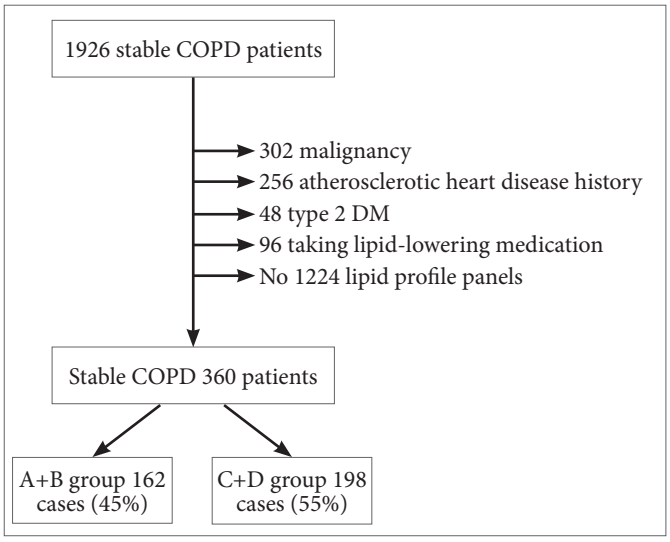

Figure. Patients flowchart

Statistical Analysis

Kolmogrow-Smirnov or shapiro-wilk test was used for the distribution of all numerical values. Categorical data were evaluated with chi-square or fisher test, if appropriate, and numerical data with student t test or man-whitney-u test, if there are 2 groups, and with Anova or kruskal wallis if there are more than two groups. Independent risk factors affecting disease severity in our 2 main groups, which were determined as A+B group and C+D group, were analyzed by multivariate regression analysis. The model fit of the created regression analysis was evaluated with the hosmer and lemeshow test, and large values above 0.05 were considered significant. For all other p values in our study, values below $p<0.05$ were considered statistically significant. Analyzes were made using theinternational bussiness machines statistical package for the social sciences (IBM SPSS) 22 program. .

RESULT

In our study, there were 360 cases, 293 (81.4%) of which were male. The mean age was 67.61±8.7 years. According to the combined COPD assessment, 37 cases (10.3%) were group A, 125 cases (34.7%) were group B, 20 cases (5.6%) were group C, and 178 cases (49.4%) were group D, while A+B group 162 was formed separately. cases (45%) and C+D group consisted of 198 cases (55%). As expected, the forced expiratory volume first second (FEV1) value was significantly lower in the C+D group ($p<0.0001$).

When potential confounding factors that may affect lipid values are excluded from the study; Other conditions such as age, gender, smoking characteristics, BMI, and hypertension were found to be similar in both study groups. Other spirometric and clinical features are in **Table 1**.

Table 1: Basal features				
Characteristics	**Total n=360**	**A+B group n=162 (45%)**	**C+D group n=198 (55%)**	**P value**
Age	67.61±8.7	67.25±8.61	67.91±8.79	P=0.47
Gender				P=0.062
Male	293 (81.4%)	125 (77.2%)	168 (84.8%)	
Female	67 (18.6%)	37 (22.8%)	30 (15.2%)	
Smoking:				P=0.089
Quitted	326 (90.6%)	142 (87.7%)	184 (92.9%)	
Still smoking	34 (9.4%)	20 (12.3%)	14 (7.1%)	
GOLD				P<0.0001
1	9 (2.5%)	8 (4.9%)	1 (0.5%)	
2	119 (33.1%)	70 (43.2%)	49 (24.7%)	
3	133 (36.9%)	53 (32.7%)	80 (40.4%)	
4	99 (27.5%)	31 (19.1%)	68 (34.3%)	
Hypertension				P=0.815
Present	56 (15.6%)	26 (16%)	30 (15.2%)	
Not present	304 (84.4%)	136 (84%)	168 (84.8%)	
VKI (kg/m²)	25.4±4.12	25.78±4.16	25.09±4.07	
<25	190 (52.8%)	86 (53.1%)	104 (52.5%)	P=0.113
≥25	170 (47.2%)	76 (46.9%)	94 (47.5%)	P=0.915
FEV1				
Liter	1.22±0.56	1.39±0.62	1.08±0.46	P<0.0001
Percent	43.75±17.86	49.33±19.12	39.18±15.35	P<0.0001
mMRC				P<0.0001
0-1	60 (16.7%)	38 (23.5%)	22 (11.1%)	
2	130 (36.1%)	86 (53.1%)	44 (22.2%)	
3	90 (25%)	33 (20.4%)	57 (28.8%)	
4	80 (22.2%)	5 (3.1%)	75 (37.9%)	
Hospitalisation/yr	0.68±1.08	0(0%)	1.24±1.19	P<0.0001
Exacerbation/yr	2.01±2.62	0.25±0.43	3.44±2.78	P<0.0001

As seen in **Table 2**, WBC, neutrophil, lymphocyte, NLR, monocytes, hemoglobin, CRP, Albumin, HDL, MHR were found to be different between the groups regarding statstical significance, while cholesterol, triglyceride, cholesterol and LDL levels were not at this level of significance.

As seen in **Table 3**; As a result of the combined COPD assessment; Independent risk factors were investigated in terms of disease stages. First, the model fit was evaluated with the hosmer and lemeshow test and it was found to be a suitable model (chi-square=6.44, p=0.598). Then, MHR, CRP and albumin values were found to be independent risk factors (**Table 3**) .

Table 3. Independent risk factors in COPD groups as a result of multivariate regression analysis

Characteristic	**B**	**p-value**	**(95% GA)**	**Odds ratio**
MHR	0.064	P<0.0001	1.029-1.104	1.066
CRP	0.137	P<0.0001	1.074-1.225	1.147
Albumin	-0.077	P=0.003	0.879-0.975	0.926

(Age, gender, smoking, hypertension, BMI, total cholesterol, triglyceride, HDL, LDL, MHR, CRP, Albumin values formed the model. The data were analyzed in terms of independent risk factors, including the dependent variable COPD group stages. The model fit was evaluated with the hosmer and lemeshow test, and it was observed as p=0.598 and chi-square test=6.44, indicating a good test fit.)

Table 2. Laboratory features

Characteristics	**Total n=360**	**A+B group n=162(%45)**	**C+D group n=198(%55)**	**Pvalue**
WBC (×10³/µL)	8.62±2.61	8.09±2.35	9.05±2.73	P<0.0001
Neutrophil (×10³/µL)	4.07±4.28	2.9±1.72	5.03±5.37	P<0.0001
Lymphocyte (×10³/µL)	1.89±0.78	2.01±0.74	1.78±0.8	P=0.006
NLR (%)	4.07±4.28	2.9±1.72	5.03±5.37	P<0.0001
Monocytes (×10³/µL)	0.66±0.30	0.58±0.24	0.72±0.33	P<0.0001
hemoglobin(g/dL)	14.06±1.99	14.33±1.81	13.84±2.11	P=0.022
CRP (mg/L)	4.26±4.25	3.07±3.09	5.22±4.81	P<0.0001
Albumin (g/L)	38.91±6.54	40.66±5.19	37.48±7.17	P<0.0001
Cholesterol (mg/dL)	187.97±46.58	192.12±43.3	184.58±48.94	P=0.126
Triglyceride (mg/dL)	127.06±74.23	126±76.38	127.94±72.61	P=0.805
HDL (mg/dL)	47.57±13.87	49.59±12.68	45.91±14.59	P=0.012
LDL (mg/dL)		117.16±37.9	114.02±38.94	P=0.441
Monosit/HDL (%)	0.015±0.011	0.012±0.006	0.018±0.014	P<0.0001

WBC=leukocytes, NLR= neutrophil lymphocyte ratio, CRP= C reactive protein, HDL=high-density lipoprotein, LDL=low-density lipoprotei

DISCUSSION

In our study, we showed that high MHR rate, high CRP and low albumin values may be independent risk factors for high disease severity in our age, sex, and BMI-matched COPD groups. COPD is a chronic disease and the expected morbidity and morbidity increase with disease severity (1).

COPD is a systemic inflammatory disease and is often associated with comorbidities, and the persistence of inflammation may also lead to other diseases (3-5). In the study of Breyer et al. (5) in which patients with COPD and healthy controls and BMI of 25 and above were evaluated, they found the prevalence of metabolic syndrome to be high in the COPD group and they found a low mean HDL level in the copd group.

Features such as obesity and systemic steroid use may affect HDL values in patients with COPD (16). In this respect, we included patients who were in the stable period and did not need steroids and were matched in terms of BMI. In our sample group, the mean BMI was 25.4±4.12 and the male ratio was 80%, which is consistent with literature data (17-19). In addition, in our study, both groups were similar in terms of gender. HDL is a lipoprotein that carries cholesterol from non-hepatic cells to the liver. Low HDL values are a significant risk factor for atherosclerosis and cardiovascular diseases. HDL molecule shows antiatherogenic activity by regulating LDL oxidation. HDL has antiatherogenic effects as well as antioxidant and anti-inflammatory effects, and low HDL is usually associated with high LDL (6,7).

Monocytes are cell types that, during inflammation, interact with platelets and endothelial cells to release pro-oxidant and pro-inflammatory cytokines, thereby causing medial damage to smooth muscle cells, leading to differentiation, apoptosis, and increased oxidative stress (8,9). For this purpose, we analyzed the lipid profile, MHR, crp, albumin and hemogram data in terms of inflammatory markers that may be associated with the severity of COPD in our study. Smoking, hypertension and BMI, which could be potential confounding factors and affect inflammation, were similar in both groups, in other words, these conditions did not affect our data. Conditions that are likely to affect other significant lipid levels, such as malignancy, type 2 diabetes, and systemic steroid use were excluded from the study (20,6).

Smoking may also negatively affect HDL levels (21), but there was no difference between the groups in terms of smoking characteristics in our study (p=0.089). Zafirova-Ivanovska B et al. (16) in their study, they found high cholesterol and LDL values and low HDL values in the very severe COPD group in the lipid profile results they evaluated in severe and very severe COPD groups, but only the high cholesterol value reached the level of statistical significance. Markelic et al. (12) In their study with COPD and healthy controls, they found high monocytes, high MHR and high HDL values in the COPD group, and they also found a relationship between high MHR rate and more airway limitation. Yakar et al. (11) In their study with COPD and healthy controls, they found MHR to be high in the COPD group, but they found that it was not associated with the severity of the disease in the COPD groups. Nillawar et al. (13) In their study with COPD and healthy controls, they found that there was no difference between lipid profile values. Can, U., Yerlikaya In their study, they found that the HDL value was lower in the COPD group compared to the healthy controls, but they found that there was no difference in LDL, triglyceride and cholesterol values (14). Sariaydin GS. (15) found in their study lower HDL, higher triglyceride and higher CRP levels in the COPD group compared to healthy controls, while they found similar cholesterol and LDL levels. When we examine the lipid profile values in our groups that we have divided as COPD A+B and C+D in our current study; We found low HDL values in the C+D group ($p=0.012$), but we found similar cholesterol, triglyceride and LDL levels in this C+D group ($p>0.05$). We found high monocytes ($p<0.0001$), low albumin ($p<0.0001$) and high CRP ($p<0.0001$) values in the C+D group. Leukocyte, neutrophil, and NLR values were also found to be high in the C+D group, which is consistent with the literature data (22, 23). When we analyzed the severity of COPD disease in our study group to examine the independent risk factors with multivariate regression analysis as seen in Table 3, we found that the statistical significance levels of high MHR rate and CRP value and low albumin values continued in the C+D group.

The main limitations of our study are that it is a single-center and retrospective study. Although the BMI was found to be similar between the groups, the fact that the nutritional habits of the cases were not known is our other limitation.

CONCLUSION

In our study, we think that high MHR rate, high CRP, and low albumin values in stable COPD patients may be a stimulant for increased disease severity. In our study, MHR, CRP and albumin values may vary in advanced COPD stages in stable COPD patients.

ETHICAL DECLARATION

Ethics Committee Approval: This study was approved by the Health Sciences University Keçiören Education and Training Hospital Clinical Studies Ethics Board (Date: 22.03.2022, Decision No: 2012-KAEK-15/2490).

Informed Consent: Since the study was designed retrospectively, informed consent was not obtained from the patients.

Conflict of Interest Status: The authors declared that there was no conflict of interest in this study.

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Author Contributions: All authors; declared that they participated in the design, execution, and analysis of the article and approved the final version.

REFERENCES

1. GOLD. Global strategy for the diagnosis, management, and prevention of chronic obstructive lung disease; 2021
2. Saymaz Z T, Çelik D, Yıldız M, Ertan Ö. The effects of D-dimer high rates on prognosis and mortality in chronic obstructive respiratory disease. Anatolian Curr Med J 2022; 4: 44-50.
3. Agust, A, Lisa D. Edwards,Stephen I, et al. Evaluation of COPD Longitudinally to Identify Predictive Surrogate Endpoints (ECLIPSE) Investigators: Persistent systemic inflammation is associated with poor clinical outcomes in COPD: a novel phenotype. PloS one 2012; 7.
4. Wang Z. L. Evolving role of systemic inflammation in comorbidities of chronic obstructive pulmonary disease. Chinese Med J 2010; 123: 3467-78.
5. Breyer M K, Martijn A. Spruit,Corrine K, et al. Prevalence of metabolic syndrome in COPD patients and its consequences. PloS one 2014; 9.
6. Assmann G, Gotto Jr A M. HDL cholesterol and protective factors in atherosclerosis. Circulation 2004; 109: 8.
7. Welty FK. How do elevated triglycerides and low HDL-cholesterol affect inflammation and atherothrombosis?. Curr Cardiol Rep 2013; 15: 1-13.
8. Ancuta P, Wang J, Gabuzda D. CD16+ monocytes produce IL-6, CCL2, and matrix metalloproteinase-9 upon interaction with CX3CL1-expressing endothelial cells. J Leukoc Biol 2006; 80:1156-64.
9. Biswas SK. Does the Interdependence between Oxidative Stress and Inflammation Explain the Antioxidant Paradox? Oxid Med Cell Longev 2016.
10. Canpolat U, Çetin EH, Cetin S et al. Association of monocyte to HDL cholesterol ratio with slow coronary flow is linked to systemic inflammation. Clin Appl Thromb Hemost 2016; 22: 476 82.
11. Yakar H I, Kanbay A. Could monocyte level/HDL cholesterol ratio predict cardiovascular diseases in patients with COPD. Niger J Clin PractNiger J Clin Pract 2020; 234: 450-5.
12. Markelić I,Iva H,,Dunja R, et al. Lipid profile and atherogenic indices in patients with stable chronic obstructive pulmonary disease. Nutr Metab Cardiovasc Dis 2021; 31: 153-161.
13. Nillawar A N, Joshi K B, Patil S B, Jayshree S, Suhas J. B. Evaluation of HS-CRP and Lipid Profile in COPD. J Clin Diag Res JCDR 2013; 7: 801.
14. Can U, Yerlikaya F H, Yosunkaya, S. Role of oxidative stress and serum lipid levels in stable chronic obstructive pulmonary disease. Journal of the Chinese Med Assoc 2015; 78: 702-8.
15. Gunay S, Sariaydin M, Acay A. New predictor of atherosclerosis in subjects with COPD: atherogenic indices. Respir Care 2016; 61: 1481-87.
16. Beti Z-I, Jagoda S, Dejan D, et al. The level of cholesterol in COPD patients with severe and very severe stage of the disease. Open access Macedonian J Med Sci 2016; 4: 277.
17. Özkan S. Risk assessment and rate of readmission within 30 days of discharge after hospitalization for acute exacerbation of chronic obstructive pulmonary disease. Tuberk Toraks 2021; 69: 328-37.
18. Melih B, Gulru P, Hulya Ş, et al. Does the benefit from pulmonary rehabilitation differ between phenotypes in chronic obstructive pulmonary disease? Eurasian J Pulmonol 2021; 23: 32.
19. Yakar HI, Kanbay A. A new predictor to determine the exacerbation and treatment of chronic obstructive pulmonary disease: eosinophil/neutrophil ratio. Tuberkuloz Toraks 2020; 68: 260-7.
20. Xilin Y, Wing YS, Ronald C, et al. Low HDL cholesterol, metformin use, and cancer risk in type 2 diabetes: the Hong Kong Diabetes Registry. Diabetes Care 2011; 34: 375-80.
21. Garrison RJ, Kannel WB, Feinleib M, et al. Cigarette smoking and HDL cholesterol the Framingham offspring study. Atherosclerosis 1978; 30: 17-25.
22. İn E, Kuluözturk M, Öner Ö, Deveci F. The importance of neutrophil-to-lymphocyte ratio in chronic obstructive pulmonary disease. Turkish Thoracic J 2016; 17: 41.
23. El-Gazzar AG, Kamel M H, Elbahnasy OKM, El-Naggar MES. Prognostic value of platelet and neutrophil to lymphocyte ratio in COPD patients. Exp Rev Respir Med 2020; 14: 111-6.

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A new era in fundamentals of bone homeostasis: biocompatibility of bone mineral doped fluoride ions with osteoblast cells in the balance of calcium and phosphate metabolism

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ABSTRACT

Aim: The use of biocompatible bone tissue grafts, filling materials, bone minerals, and implants, particularly in medicine and dentistry studies, has expanded significantly in recent years, as have expectations from the materials. We aimed to test the biocompatibility and wound and tissue biocompatibility of many grafts and similar materials used in medicine and dentistry with tests such as cytotoxicity, scratch assay, cell adhesion, and hemolysis.

Material and Method: In this study, the interaction of fluorine ions with a dental material was investigated by biological activity experiments. In addition, studies were carried out on important osteoblast cells for tissue regeneration control. For this process, cell migration analysis, which we do not encounter frequently in the literature, was used to examine the interaction of cells with biomaterials more sharply.

Results: Flor ions do not create a cytotoxic effect and also increase the viability of osteoblasts which is important for tissue regeneration and are bone precursor cells.

Conclusions: In this study, in which the efficiency of osteoblast cells was discussed, it was concluded that 2% fluorine added material had more effective biological results compared to the increase in fluorine ion ratio.

Keywords: Tooth minerals, hydroxyapatite, osteoblast, cell migration, hemolysis

INTRODUCTION

The use of biocompatible bone tissue grafts, filling materials, bone minerals and implants, especially in dentistry studies, has increased considerably in recent years and the expectations from these materials have increased proportionally (1,2). These grafts, which are used to repair and regenerate bone tissue; It is expected to have a structure that is biocompatible, has a cell viability-enhancing effect, has high mechanical strength, and triggers osteogenesis, cementogenesis and functional periodontal ligament formation (2). Bone grafts, which are used both in dentistry and in many areas of medicine, are divided into many groups as autogenous bone allografts, xenografts and alloplastic grafts in order to induce bone tissue regeneration or to make implants more compatible. Hydroxyapatites, the primary mineral compound of bone, which is in the alloplastic group and whose use has increased in recent years, is the group that provides advantage in its design and application with its compatibility and positive results with different mineral additives (3-7). In the process of bone fractures, injuries and regeneration; The osteoinduction process is the process of differentiation of mesenchymal stem cells into osteoblasts, one of the bone precursor cells, and thus, osteoblast cells in the bone graft provide a three-dimensional skeleton for vascularization and cell migration. As the rate of osseointegration, which is a direct structural and functional link between living bone and the surface of the designed implant or graft, increases, the biological compatibility also increases (8). Osteogenesis occurs by mesenchymal stem cells and their differentiation by osteoblasts, and thus bone formation is stimulated (9).

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While there are osteoblasts, osteoclasts, and bone lining cells outside the bone tissue, there are osteocytes in the mineralized interior (10). They are fully differentiated cells responsible for the production of the bone matrix, secreting collagen type I and non-collagenous proteins of the bone matrix by osteoblast cells. Osteoblasts are also extremely important cells for bone, responsible for the production of factors that affect the differentiation and function of osteoclasts (11). Hydroxyapatite, which is frequently preferred in alloplastic grafts with hydroxyl ions (OH-) in its chemical structure and consisting of high carbonate ions, also serves as a three-dimensional skeleton in dentin tissue regeneration and repair (12). Increasing its effectiveness with different additives shows that it can be used in different areas (13). In this study, Hydroxyapatite was added with fluoride and its compatibility with cells, its effect on cell migration, and its biocompatibility for human use were evaluated. For this purpose, cell viability analysis according to the 10993-5 standard, cell migration to evaluate the progression potential of cells, and hemolysis tests to evaluate their interaction with blood was performed. In addition, the adhesion of the cells to the biomaterial surface was examined with a scanning electron microscope.

MATERIAL AND METHOD

Chemicals and Reagents

Osteoblast cell, 25 cm^2 and 75 cm^2 cell culture flasks, 96 well plates, serological pipettes and other plastic materials (Corning, NY, USA). DMEM, Dulbecco Modified Medium (DMEM) L-glutamine, Penicillin-streptomycin (PS), fetal bowine serum (FBS), and Trypsin-EDTA (Biological Industries Ltd. Kibbutz Beit Haemek, Israel).

Characterization of F- Doped HAp

Cell culture: After the cells previously frozen in the liquid nitrogen tank were thawed at 37°C in a short time, they were transferred to a 15 mL falcon tube in a sterilized Class II cabinet, and some medium was added. The prepared falcon was centrifuged at 2500 rpm for 2 minutes to allow the cells to collapse. After the supernatant was discarded, 3 mL of DMEM medium (containing 10% FBS, 1% PS) was added to the falcon, and after it was homogenized, it was cultivated into 25cm^2 and 75cm^2 flasks. The cultivated flasks were incubated at 37 °C in a 5% CO_2 incubator. After the cells were 80% confluent, they were passaged at least three times and prepared for the MTT cytotoxicity test and wound healing tests. After cell analysis, osteoblast cells were implanted on the surface of the material and the frequency of cell attachment was tried to be determined by imaging with a scanning electron microscope (14).

Preparation of extracts: EN ISO 10993-12 is used for extraction of medical devices and biomaterials. In this way, it is known that the properties of the materials pass into the extraction liquid when the materials that cannot be applied directly on the cell come into contact with suitable liquids at suitable temperatures under certain conditions. Extraction of test materials as specified in ISO 10993-12, the guideline for biological characterization testing of medical devices and materials requiring biocompatibility; It was incubated at 37°C with 1X DMEM for 24 hours, corresponding to 0.1 g/mL. Then, a suitable sterilization was performed to remove the possibility of possible contamination and material residue and to remove it from the environment. 10% FBS, 1% ps (penicillin/streptomycin) and 1% L-glutamine were added to the extract medium and made ready for testing. The prepared extract was used within 24 hours. The prepared extract directly represents the sample according to the standard and is named as 1/1(full) concentration.

Cytotoxicity assay: After the cells previously frozen in the liquid nitrogen tank were thawed at 37°C in a short time, they were transferred to a 15 mL falcon tube in a sterilized Class II cabinet, and some medium was added. The prepared falcon was centrifuged at 2500 rpm for 2 minutes to allow the cells to collapse. After the supernatant was discarded, 3 mL of DMEM medium (containing 10% FBS, 1% PS) was added to the falcon, and after it was homogenized, it was cultivated into 25 cm^2 and 75 cm^2 flasks. The cultivated flasks were incubated at 37 °C in a 5% CO_2 incubator. After the cells were 80% confluent, they were passaged at least three times and prepared for the MTT cytotoxicity test and wound healing tests.

Scrach assay (cell migration): This test method is a useful in vitro method to detect cell migration. Cell migration and spreading abilities of osteoblast cells were evaluated using a scratch wound assay that measures the expansion of a population of cells on surfaces. Cells were seeded in 24-well cell culture dishes at 2.5×105 cells/mL, incubated at 37 °C for 24 hours. After the cells were confluent and covered the plate, a linear wound sample was created on the layer in one go with a sterile 100 µl plastic pipette tip, the plates were washed 2 times with PBS (phosphate buffered saline) to remove the raised cells and media residue. As the control group, only the medium prepared with DMEM and the extract of the material (1/1 (100%)) as the test substance were applied and incubated at 37 °C containing 5% CO_2. While the created scar was incubated for 24 hours, the wound areas were photographed at 0, 12 and 24 hours.

Hemolysis test: To assess the blood compatibility of the samples in the test groups, they were incubated in a tube with Mg/Ca Free PBS at 10.2 g/mL for 24 hours below 37 °C as specified by ISO-10993-12. Equal volumes of blood were collected from 3 donors in 0.13 M citrate tubes and 7 mL of extraction liquid for each test sample was divided

into tubes. The standard curve, the constant F (calibration coefficient) was determined for all products by dilution of the hemoglobin standard. The required hemoglobin concentration for the test is specified as 10±1 in the standard. In the present experiment, the concentration value of hemoglobin was calculated as 9.8. For each sample, 7 mL of extract was added to 1 mL of blood and incubated at 37°C for 3 hours. Blood and control groups interacting with the sample were centrifuged at 700-800 G for 15 minutes. The supernatant plasma and drabkin are mixed at a ratio of 1:1 and measured in a spectrophotometer at 540 nm after 15 minutes of incubation (15).

Statistical Analysis

All experiments were performed in triplicate. Data were analyzed using one-factor analysis of variance to determine equality of population means and Student's t-tests were performed between respective populations where $P< 0.05$ was considered statistically significant.

RESULT

Biological Activity

Biomaterials, which enable living tissues in the human body to perform their functions or restructure, are used not only as implants and prostheses, but also in extracorporeal devices, diagnostic purposes, and wound treatment (16). Some biomaterials are also included in the medical device class, and in order for these biomaterials to be suitable for human use, they should not be toxic and carcinogenic at first, depending on the characteristics of the target area to be used; it should be mechanically durable and non-corrosive and should not cause reaction other than routine reactions of the body (17). For all these requirements, there is the "EN ISO 10993-1: Biological Evaluation of Medical Devices Part 1- Evaluation and testing" standard, which is accepted in all countries of the world. In our study, we carried out biological activity tests by taking this standard and the standards it references as a guide.

Cytotoxicity Assay

The most important parameter for the use of the material is its suitability for human use, and in vitro cytotoxicity testing under laboratory conditions is the first and most important step for this suitability. In this experimental system designed on the effect of drugs or materials applied on cells on cell viability, application materials that do not affect the viability of cells, do not reduce them below 70% or increase viability are found suitable for use in vitro. In this test method, "EN ISO 10993-5: Extracorporeal cytotoxicity tests" standard was taken into consideration. In its qualitative evaluation, the effect of extracts on cells was examined microscopically. MTT cytotoxicity assessment was done both quantitatively and qualitatively. Qualitative evaluation results are indicated in **Table 1**. Quantitative evaluation results are shown in **Table 2**. Calculation of the % vitality was made using the equation below.

Table 1. Qualitative morphological grading of the cytotoxicity of the extracts

Test material	Reaction	Status of cultures
Negative control	0	Discrete intra-cytoplasm granules, no cell destruction, no decrease in cell proliferation
Positive control	4	All or almost all of the cell layers were destroyed
1 F (1/1)	0	Discrete intra-cytoplasm granules, no cell destruction, no reduction in cell proliferation
2 F (1/1)	0	Discrete intra-cytoplasm granules, no cell destruction, no reduction in cell proliferation
3 F (1/1)	0	Discrete intra-cytoplasm granules, no cell destruction, no reduction in cell proliferation

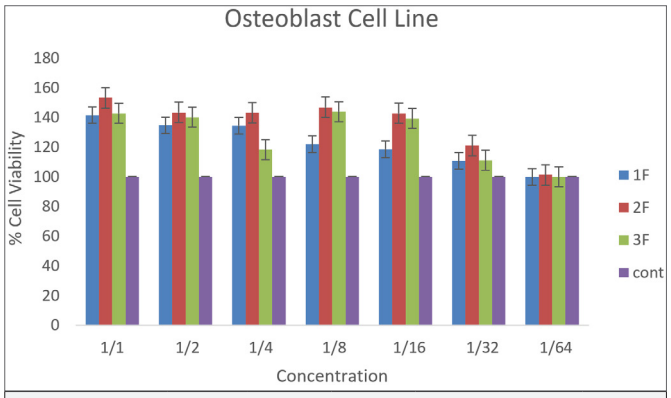
Table 2. %Hemolysis values of 1F, 2F and 3F samples after application

Groups	Abs (540 nm)	SHC	% hemolys	Color	Clarity	Particulates present
Negative control	0.0474	0.0267	0.2645	Colorless	Clear	No
1 F extract	0.0482	0.0271	0.2710	Colorless	Clear	No
2 F extract	0.0545	0.0298	0.3018	Colorless	Clear	No
3 F extract	0.0508	0.0267	0.2921	Colorless	Clear	No
1 F direct contact	0.0483	0.0267	0.2605	Colorless	Clear	No
2 F direct contact	0.0552	0.0327	0.3176	Colorless	Clear	No
3 F direct contact	0.0496	0.0293	0.2774	Colorless	Clear	No
Positive control	1.7018	0.1927	18.672	Colorless	Red	No

Cell viability %=Optical Density, OD570 sample/Optical Density, OD570 (control) ×100

Accordingly, while the negative control did not show any toxic effect on cells (0), natural rubber used as a positive control showed a high degree of toxicity as expected (4). When the cytotoxic effect of the sample extracts was examined, it was evaluated as zero because it was not seen as toxic (0). There was no decrease in cell destruction and cell proliferation. All details can be seen in **Table 1**. According to the standard used, a rating value greater than (2) as indicated in **Table 1** is considered a cytotoxic effect. "TS EN ISO 10993-5 /C MTT Cytotoxicity Test" is used as a quantitative evaluation method and the results are evaluated statistically. **Figure 1** also shows the results obtained from the negative and positive controls used, and it is seen that the test validity criteria are met. In this experiment, the effects of 1/1 to 1/64 dilutions of the sample extract on osteoblast cells were investigated. The viability obtained from the 1, 2 and 3 % F doped samples were determined as 141.45±1.46, 153.30±4.49 and 142.82±6.46 for osteoblast cells, the immature cell type (bone cell precursor), with complete

dilution (1/1) of the samples. Each sample was seeded to evaluate the osteoblast cell adhesion status. As a result of this cultivation process, cell entities were examined by scanning electron microscopy. The visuals of the analysis in which the status of the cell assets according to the contribution rate are evaluated are shown in **Figure 2**.



Sample Concentration	1F		2F		3F	
	%	±	%	±	%	±
1	141.45	1.46	153.30	4.49	142.82	6.46
1/2	134.59	3.25	143.27	1.93	140.09	1.49
1/4	134.51	5.51	143.13	1.19	138.20	1.75
1/8	122.01	2.41	146.74	3.60	143.89	6.18
1/16	118.66	1.55	142.72	1.84	139.16	4.74
1/32	110.81	0.13	121.18	4.05	111.04	1.16
1/64	99.86	5.08	101.26	1.22	99.91	3.03

Figure 1. Percent cell viability value graphs of Osteoblast cells after 24 hours of application. The application was made between 1/1 (full concentration) and 1/64 concentration range

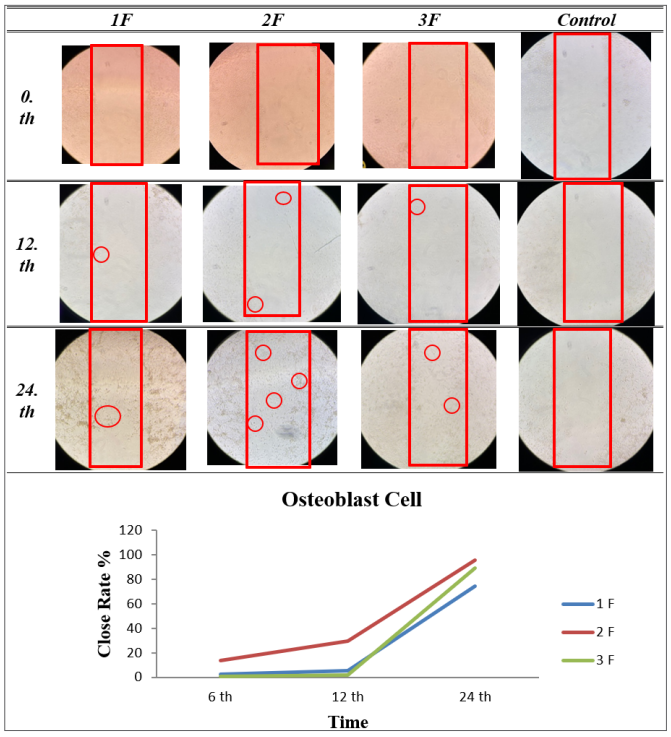


Figure 2. Fluoride-doped cell migration and wound closure analysis on osteoblast cell. (The areas marked at 0 hour are the areas where the wound was formed, and the areas where the cells 12 and 24 were migrated and the wound was closed are shown in the circle.)

Osteoblast cells, which are the immature support cells that are the precursors of the bone cell, are the cell group preferred in this study because of their potential to transform into immature bone cells and their high metabolic activity. Osteoblast cells are involved in the formation and regeneration of bones (18-21).

Osteoblasts are bone cells that are primarily responsible for synthesizing bone matrix proteins and minerals during early bone formation in the embryo, but also control bone formation and mineralization throughout life. Since they are located in areas of high metabolism where new bone formation occurs, the results of osteoblast cells were carefully examined in the study content. The cell analysis results obtained according to all three F ratios were found to be well above the 70% value defined for viability in the EN 10993-5 standard. This result showed that it is a product that supports cell proliferation effectively and has been interpreted as producing a matrix covering the old bone surface, leading to the formation of new bone cell layers and mineralization of the bone to regulate the balance of calcium and phosphate ions in the developing bone.

The increase in osteoblast cell presence increases both maturation and mineralization of the extracellular matrix. When the cell viability values were carefully examined, it was concluded that the cell viability values of the 2% F added sample had a statistically significant difference. As observed in a previous study, in the fluoride doped Hydroxyapatite medium, cell proliferation increased according to the doping ratio and decreased after the critical value. Similarly, the rate of 2% was determined as the most effective rate in this study. This finding helped us to conclude that the produced biomaterial could be well correlated with the feasibility study. According to the viability values obtained, a significant increase in cell viability and a decrease after a certain value were obtained with the increase in fluoride supplementation. The common result for all three contribution ratios is positive contribution to cell proliferation. Consistent with a study by Erdem et al. (7) it was concluded that fluoride-doped HAp ceramics did not cause toxic effects. At the same time, it is thought that these different ratios determined in the study content are sufficient values to examine the effect on the differentiation of primary osteoblast cells.

According to the contribution ratio, different results were obtained in osteoblast cell viability values. Although the cell viability value seems quite good in all three values, this value is at a better point for 2%. This shows that 2% F additive will contribute more to the remineralization of demineralized bone. Increasing the F ratio improved the osteogenic potential of these samples, as expected.

Hydroxyapatite, which has been used for bone development in dentistry and orthopedics for many years, is an inorganic component in bones and teeth and has a very similar structure to biomineralized tissues (22). It is stated that especially ion-added coatings are a good alternative to reduce implant infections. In addition, it is suggested that hydroxyapatite coatings containing fluoride, silver, etc. ions on implants can effectively inhibit bacterial adhesion and growth without affecting the activity of osteoblasts and epithelial cells (23-25). The cell study results obtained from the product in question help to conclude that hydroxyapatite will be an effective dental treatment material when its known antibacterial properties are considered. The test results show that the synthesized 1.2% and 3% F doped materials are biocompatible. It was also found that the test material showed no toxic effects on osteoblasts (bone precursor cells). The test results show that there will be no toxic effects if the sample is used in the body, or if it is broken/cracked out from the area to be applied, or if it is used as a drug synthesis substance.

The periosteum is a thick layer that surrounds the bone from the outside and is rich in collagen fibrils and fibroblasts. The inner layer contains osteoprogenitor cells (26). In the test results, it was determined that this structure of bone tissue increased the viability of 2% F material by 50% more in addition to normal proliferation. This means that the synthesized material can heal quickly on the outside in the repair of bone tissue, due to the rapid adaptation and support to the bone. Osteoblasts are present in 4-6% of bone tissue (27,28). Cytotoxicity results show that the material in the osteoblast is non-toxic, does not inhibit viability, and causes excessive proliferation. Thus, if the material is applied, osteoblasts will continue their normal biological functions and will not disrupt their natural structure.

Ramises et al. (29) and Chen et al. (30) reported that the use of titanium implants coated with HA had a viability-enhancing effect, while HA nanoparticles did not have a negative effect on cell viability. has also been shown by. In our study, HA did not have a cytotoxic effect and increased cell viability with fluoride supplementation. This shows that HAs doped with fluoride can also be used in different fields.

Student's t-tests were conducted between populations of interest, with $P<0.05$ considered statistically significant. Statistical significance for 1/1 concentration for cell line written in **Table** shown together with **Figure 1**'s captions. P value; P 0.213929711>α (0.05) (1F-2F), 0,020677283 <α (0.05) (1F-3F) and 0.123522342>α (0.05) (2F-3F). According to or by means of statistics, the most significant increase in cell viability was at 2% F.

Scratch Assay (Cell Migration) Test

One of the most important steps in wound healing is the migration of cells to the wound area, proliferating and initiating tissue restructuring. Instead of in vivo methods for wound healing, in vitro methods are more preferred in the early stages of synthesis in terms of low cost, animal welfare and fast results. This method is based on the creation of an artificial cavity by scraping on the confluent cell monolayer. The cells remaining at the edge of the wound cavity due to the effect of the applied drugs or materials move towards the opening to close that gap (in vitro wound model) and have the capacity to close the opening. The method is simple to implement and offers several advantages: (I) it mimics cell migration to some extent in vivo, and (II) is suitable for studying the regulation of cell migration by the extracellular matrix (ECM) and cell-cell interactions (31). The size of the wound cavity formed is scaled by SEM and the migration of cells into the cavity is recorded. In vitro wound healing and cell migration were calculated by measuring the areas from which field images were taken, using ImageJ, an open source image processing program. Calculations for osteoblast (**Figure 2**) cell groups are shown in detail.

The visuals and occlusion rates obtained as a result of the cell migration study performed in the light of these data are shown in **Figure 2**. Closure rates were recorded at the end of the 6, 12 and 24 hour periods. It was observed that for the best closure rate at these times, it clearly belonged to 2% F. The values obtained for 1 and 3 % F were recorded similarly to each other. At the end of the 24-hour period, a significant difference is obtained in each of the 3 additive ratios compared to the control group, but the 2% F closure rate in the 6 and 12 hour periods is remarkable. Considering the importance of the first 6 hours for compliance in dental operations, the presence of cells in the first hours is important. For this reason, the result obtained in the first 6 hours was considered valuable and it was concluded that the 2%F ratio was more effective. The capacity of filling the gap and cell migration in Osteoblast cells of the 2% F doped test samples improved compared to the control group. It was also observed that after 24 hours the cells became identical to the non-wound areas (their original state). Although the cell migration of the 1% and 3% F-added test materials also improved compared to the control group, it was found that the increase did not make a statistical difference for the two of them, but increased continuously compared to the control group. This indicates that the applied samples induce cell migration. Osteoblasts have Platelet Derived Growth Factor (PDBF) receptors, thus playing a role in cell division and chemotaxis of applied materials (32). It is thought that the finding of in vitro wound closure and cell migration of the applied materials is by this mechanism. Considering the developments in osteoblast cells and

wound healing potential of the materials planned to be used in bone tissue treatment, the promising potential of the examined materials in bone damage can be mentioned. The fact that the material has wound closure potential shows that both the cells adapt to the material and the effectiveness of the material is fast. It is predicted that if it is used in the clinic for patient health and comfort, it will save time and provide success in treatment.

Cell Adhesion

It is extremely important that the materials to be placed in the body can interact with the tissues, establish a bond with the tissue area where they are implanted, and be integrated. The adhesion of the cells to the prepared biomaterials causes the formation of cell layers and their integration with the tissue over time. Thus, it will begin to participate in the normal flow of the body as well as receive support from the material. Although cell adhesion or non-adhesion differs according to the area planned to be used, the interaction of the material with the cell will contribute to the bone tissue in our study. For this reason, cell adhesion to materials is one of the important points. Since non-biocompatible materials are generally not preferred by cells and cause cell death in contact, the adhesion of cells increases the probability that a material that is biocompatible in vitro will also be compatible in in vivo tests. It is planned that the synthesized fluoride added materials will be used to eliminate and treat bone tissue damage. For this reason, with the cell adhesion test; Scanning Electron Microscope (SEM) examination was performed as an indicator of cell morphology status and evaluation of proliferation potential. Osteoblasts grown on fluoride-doped hydroxyapatites are morphologically diverse, usually cubic, round or flat, cylindrical, and 20~50 μm in diameter. Cell sizes in the images obtained in SEM analysis are around 10-40 μm (**Figure 3**). The growing osteoblasts appeared to form a thin, single layer of squamous cells on the surface of the biomaterial. At the same time, it was observed that highly fibroblast-like and partially round cells were attached to the material by cytoplasmic extensions. As such, osteoblasts have a dendritic and flat morphology. Scanning electron microscope images also show close intercellular contacts. It shows that two cells are interconnected to form a dense layer of flat and spinocellular cells (33). Cell studies, as well as the antibacterial properties of hydroxyapatite, the presence of extracellular matrix proteins and a growth image helped us to demonstrate its osteoinductive and osteoconductive properties well (34). Although the cell presence observed in this study is considered promising for the formation of bone nodules, it does not preclude the need for future in vivo studies to support this assessment. Cell presences were seen in each of the images obtained from osteoblast cells grown on these three biomaterials. Consistent with the cytotoxicity results, cell adhesion was evaluated to be higher at the 2% additive rate compared to the additive rate. This study evaluated the biocompatibility and mineralization ability according to the increase in fluoride additive ratio. According to the results obtained, although all three biomaterials were evaluated as suitable as hard tissue repair material, it was concluded that the cells were overexpressed at the 24th hour of the experiment, especially in the areas where 2% F added samples were applied. This suggests that if the implant to be applied to the bone tissue is covered with material, it will result in an increase in bone precursor cells and the corresponding implant will provide a rapid adaptation to the body. It is also promising because the cytoskeleton prepared with 2% F doping level for bone tissue engineering will adapt quickly and support the formation of new tissue.

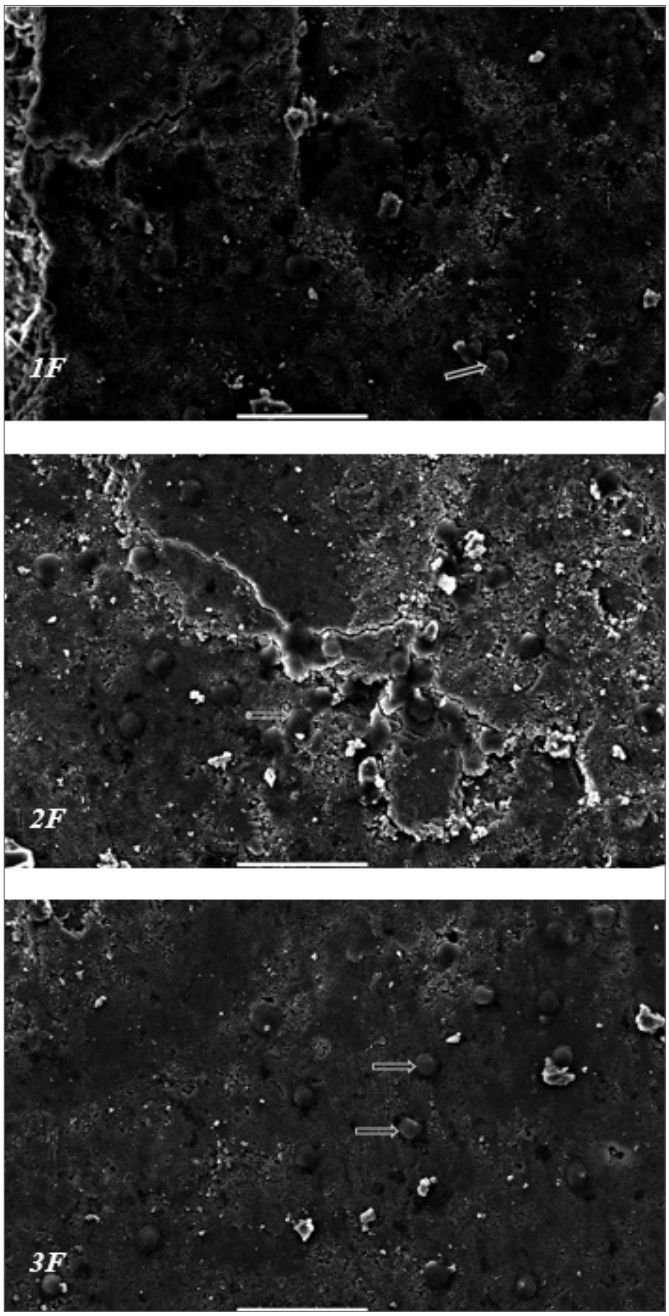


Figure 3. SEM images of fluorinated materials on which cells were applied (arrows indicate adherent cells). (Scale bar 50μm)

Hemolys Test

After the medical device or material is produced, the material must be subjected to some tests according to its intended use. In order to ensure the safety of the material used in devices that come into contact with blood (such as vascular prostheses), the compatibility of the synthesized material with blood should be evaluated. Hemolysis testing is required for all devices and device materials, except for materials that come into contact with intact skin and/or mucous membranes. The general principle of the test is to measure the damage of red blood cells exposed to the substance. Although there are many studies on the mechanism of hemolysis, there is no clear information on how exactly it occurs (35). For this reason, if the material to be used affects the mechanism hemolytically, it is not possible to intervene and repair it. The breakdown of red blood cells in the blood can cause both the normal flow of blood to change and the increase in erythrocyte production in the bone marrow, leading to enlargement of the bone marrow. For all these reasons, it is desired that the materials to be applied should not have a hemolytic effect. The application steps of the test as well as the material and method are specified in the 8th article. The test method applied is "TS EN ISO 10993-4 Biological evaluation of medical devices - Part 4: Selection of blood interaction tests" standard. It is valid because the test acceptance criteria specified in the standard are met. The experimental results obtained show that the sample does not have a hemolytic effect due to the absence of a value greater than 5% at the Hemolytic index levels given in **Table 1**. has been tested and the test results reveal that no hemolytic effect was observed in either case. However, in such a case, it is recommended to perform other (blood-interacting) tests such as coagulation, platelet, hematology and complement. The hemolytic index calculation results are tabulated in **Table 2**, and it was determined that both the extracts and the direct interactions with the blood of HAp samples with F partial substitutes produced in this study did not show any hemolytic effect in both cases (**Table 2**). In this way, this material does not show a hemolytic effect in case of any interaction with the blood in the targeted area. However, in such a case, it is highly recommended to perform other tests.

CONCLUSION

When the biological activity tests were evaluated, it was observed that the results of all tests (cytotoxicity, scratch assay, cell adhesion and hemolysis) were consistent with each other. It has been determined that the applied materials do not have a cytotoxic effect, and they also support the increase of the viability of osteoblasts, which are bone precursor cells. In particular, it was determined that 2% F has no toxic effect and increases cell viability by 50%, the rate of wound closure in the scratch assay test is higher than other materials, and it tends to close the area completely after the 12th hour and supports the complete closure of the wound area at the end of 24 hours. 1, 2, 3 F added materials have no hemolytic effect; indicates the suitability of contact with the skin or blood vessels. It is found that 2% F is suitable for use in materials evaluated in vitro. In order for human use to be possible, in vivo studies in which all body system functions can be evaluated, and cell increase and growth factors such as PDBF (Platelet Derived Growth Factor), EGF (Epidermal Growth Factor), FGC (Fibroblast Growth Factor), VEGF (Vascular Endothelial Growth Factor) It is recommended to examine the effect on support elements such as collogen type I and collogen type III.

ETHICAL DECLARATIONS

Ethics Committee Approval: Ethics committee approval is not required for this study.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

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Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

REFERENCES

1. Brunsvold MA, Mellonig JT. Bone grafts and periodontal regeneration. Periodontol 2000 1993; 1: 80-91.
2. Singh J, Takhar RK, Bhatia A, Goel A. Bone graft materials: dental aspects. J Nov Res Healthc Nurs 2016; 3: 99-103.
3. Poundarik AA, Wu PC, Evis Z, et al. A direct role of collagen glycation in bone fracture. J Mech Behav Biomed Mater 2015; 52: 120-30.
4. Evis Z, Doremus RH. A study of phase stability and mechanical properties of hydroxylapatite–nanosize α-alumina composites. Materials Science and Engineering C 2007; 27: 421-5.
5. Wang H, Li Y, Zuo Y, Li J, Ma S, Cheng L. Biocompatibility and osteogenesis of biomimetic nano-hydroxyapatite/polyamide composite scaffolds for bone tissue engineering. Biomaterials 2007; 28: 3338-48.
6. Liu C, Wang W, Shen W, Chen T, Hu L, Chen Z. Evaluation of the biocompatibility of a nonceramic hydroxyapatite. J Endod 1997; 23: 490-3.
7. Baglar S, Erdem U, Dogan M, Turkoz M. Dentinal tubule occluding capability of nano-hydroxyapatite; The in-vitro evaluation. Microsc Res Tech 2018; 81: 843-54.
8. Khan SN, Cammisa FP Jr, Sandhu HS, Diwan AD, Girardi FP, Lane JM. The biology of bone grafting. J Am Acad Orthop Surg 2005; 13: 77-86.
9. Sukumar S, Drízhal I. Bone grafts in periodontal therapy. Acta Medica (Hradec Kralove) 2008; 51: 203-7.

10. Aubin JE, Triffitt JT. Mesenchymal stem cells and osteoblast differentiation. In Principles of Bone Biology, Academic Press 2002; 59-81.
11. Sandberg MM. Matrix in cartilage and bone development: current views on the function and regulation of major organic components. Ann Med 1991; 23: 207-17.
12. Teruel Jde D, Alcolea A, Hernández A, Ruiz AJ. Comparison of chemical composition of enamel and dentine in human, bovine, porcine and ovine teeth. Arch Oral Biol 2015; 60: 768-75.
13. Kim HW, Knowles JC, Li LH, Kim HE. Mechanical performance and osteoblast-like cell responses of fluorine-substituted hydroxyapatite and zirconia dense composite. J Biomed Mater Res A 2005; 72: 258-68.
14. Dinc DO, Yilmaz M, Sebnem Cetin S, Turk M, Piskin E. Gold-nanoisland-decorated titanium nanorod arrays fabricated by thermal dewetting approach. Surface Innovations 2019; 7: 249-59.
15. Jenifer A, Senthilarasan K, Arumugam S, Sivaprakash P, Sagadevan S, Sakthivel P. Investigation on antibacterial and hemolytic properties of magnesium-doped hydroxyapatite nanocomposite. Chemical Physics Letters 2021; 771: 13853.
16. Park J, Lakes RS. Biomaterials: an introduction. Springer Science & Business Media 2007.
17. Silver F. Biomaterials, Medical Devices and Tissue Engineering: An Integrated Approach: An Integrated Approach. Springer Science & Business Media 1993.
18. İnsal B, Pişkin İ. Kemik dokusunun fizyolojisi. Etlik Veteriner Mikrobiyoloji Derg 2017; 28: 28-32.
19. Kong YY, William JB, Penninger MJ. Osteoprotegerin ligand: a regulator of immune responses and bone physiology. Immunol Today 2010; 21: 495-502.
20. Lanyon LE. Osteocytes, strain detection, bone modeling and remodeling. Calcif Tissue Int 1993; 53: 102-7.
21. Leibbrandt A, Penninger JM. RANK/RANKL: regulators of immune responses and bone physiology. Ann N Y Acad Sci 2008; 1143: 123-50.
22. Türköz MB, Erdem Ü. Katkısız ve Gümüş Katkılı Hidroksiapatitin Fiziksel ve Kimyasal Karakterizasyonu. Int J Engineer Res Develop 2019; 11: 643-56.
23. Erdem U, Turkoz MB. Silver release of Ag (I) doped hydroxyapatite: In vitro study. Microsc Res Tech 2019; 82: 961–71.
24. Uysal I, Severcan F, Tezcaner A, Evis Z. Co-doping of hydroxyapatite with zinc and fluoride improves mechanical and biological properties of hydroxyapatite. Progress in Natural Science: Materials International 2014; 24: 340-9.
25. Erdem, U., & Turkoz, M. B. La3+ and F– dual-doped multifunctional hydroxyapatite nanoparticles: Synthesis and characterization. Microscopy Research and Technique 2021; 84: 3211–20.
26. Topaloğlu U. , Ketani M. A. , Güney Saruhan B. Kemik Doku ve Kemikleşme Çeşitleri. Dicle Üniv Vet Fak Derg 2017; 10: 62-71.
27. d'Aquino R, Graziano A, Sampaolesi M, et al. Human postnatal dental pulp cells co-differentiate into osteoblasts and endotheliocytes: a pivotal synergy leading to adult bone tissue formation. Cell Death Differ 2007; 14: 1162-71.
28. Venugopal J, Vadgama P, Kumar TS, Ramakrishna S. Biocomposite nanofibres and osteoblasts for bone tissue engineering. Nanotechnology 2007; 18: 055101.
29. Ramires PA, Romito A, Cosentino F, Milella E. The influence of titania/hydroxyapatite composite coatings on in vitro osteoblasts behaviour. Biomaterials 2001; 22: 1467-74.
30. Chen L, Mccrate JM, Lee JC, Li H. The role of surface charge on the uptake and biocompatibility of hydroxyapatite nanoparticles with osteoblast cells. Nanotechnology 2011; 22: 105708.
31. Lodish H, Berk A, Kaiser CA. et al. Moleküler hücre biyolojisi. Çeviri editörleri: Geçkil H, Özmen M, Yeşilada Ö. Palme Yayıncılık 2011; 6: 248-9.
32. Hughes FJ, Aubin JE, Heersche JN. Differential chemotactic responses of different populations of fetal rat calvaria cells to platelet-derived growth factor and transforming growth factor beta. Bone Miner 1992; 19: 63-74.
33. Fuss M, Ehlers EM, Russlies M, Rohwedel J, Behrens P. Characteristics of human chondrocytes, osteoblasts and fibroblasts seeded onto a type I/III collagen sponge under different culture conditions. A light, scanning and transmission electron microscopy study. Ann Anat 2000; 182: 303-10.
34. Korkusuz F, Korkusuz P. Calcium hydroxyapatite ceramics in orthopedic surgery. Acta Orthopaedica et Traumatologica Turcica 2006; 31: 63-7.
35. Baumann E, Stoya G, Völkner A, Richter W, Lemke C, Linss W. Hemolysis of human erythrocytes with saponin affects the membrane structure. Acta Histochem 2000; 102: 21-35.

Original Article

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Outcomes of patients with COVID-19 pneumonia treated with moderate and high dose corticosteroids

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ABSTRACT

Introduction: The mortality related to coronavirus disease-2019 (COVID-19) develops due to hyper immune response in most of the patients. The use of corticosteroids (CS) is reported to be effective in decreasing mortality and maintaining a better prognosis but the results of some studies are against the use of CS due to delayed virus clearing, adverse effects, and insignificant effect on clinical course and outcome. We aimed to evaluate the effect of CS use in hospitalized COVID-19 pneumonia patients on clinical course and mortality.

Material and Method: Demographic and laboratory data, history for CS treatment, need for oxygen support, duration of hospitalization and/or ICU follow up, and mortality of inpatients with COVID-19 pneumonia treated between 15.03.2020 and 15.06.2021 at Sultan 2. Abdülhamid Han Training and Research Hospital were collected. The whole cases were divided into two groups as CS group (who were given CS) and the control group (who did not receive CS). All parameters were compared between the two groups.

Results: A total of 185 patients (122 CS group and, 63 control group) were included in the study. The patients in the CS group were younger than the controls (median age was 63 (30-91) years and 71 (34-91) years respectively, p<0.001). In the CS group compared to controls, the number of cases followed in ICU and intubated was lower (55 [45.1%] cases vs. 53 [84.1%] cases, p<0.001 and 40 [32.8%] cases vs. 53 [84.1%] cases, p<0.001, respectively) but, the time for ICU and hospital stay was longer (13 [1-32] days vs.6 [1-29] days, p<0.001 and 11 [5-44] days vs. 9 [4-35]days, p=0.005, respectively). The number of cases who died was less in the CS group compared to controls significantly (35 [28.7%] cases vs. 53 [84.1%] cases respectively, p<0.001).

Conclusion: CS treatment may be related to better prognosis and less mortality in COVID-19 pneumonia inpatients although it may prolong the time for ICU follow up and hospitalization.

Keywords: COVID-19, pneumonia, corticosteroid, mortality

Our research's data was presented in 9. International Medicine and Health Sciences Congress as 'Oral Presentation' on March 2022.

INTRODUCTION

The mortality due to coronavirus disease-2019 (COVID-19) was explained by multi-organ failure developed as a result of severe hyper immune response named "cytokine storm" in most of the patients (1,2). The idea for the use of corticosteroids (CS) in management of hospitalized COVID-19 patients was supported by the information from previous studies that it might have beneficial effects in overcoming both hyper inflammation and adult respiratory distress syndrome (ARDS). The use of CS is reported to be effective on decreasing mortality and resulting in a better outcome (3-10). In contrast, some studies reported that CS use was associated with delayed virus clearing and did not improve survival, reduce hospitalization duration or intensive care unit (ICU) admission rate and/or use of mechanical ventilation (11,12). In this retrospective, observational study we aimed to evaluate the effect of CS use in hospitalized COVID-19 pneumonia patients on clinical course and mortality.

MATERIAL AND METHOD

Adult inpatients aged over 18 years, treated for COVID-19 pneumonia in the Sultan 2. Abdülhamid Han Training and Research Hospital between 15.03.2020 and 15.06.2021 were included in the study. COVID-19 pneumonia was diagnosed based on polymerase chain reaction (PCR) positivity in nasopharyngeal swab or lower respiratory tract samples and radiological findings consistent with COVID-19

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pneumonia on chest computed tomography (CT). This study was approved by Ümraniye Training and Research Hospital Clinical Researches Ethics Committee (Date: 27.05.2021, Decision No: B.10.1.TKH.4.34.H.GP.0.01/179). All procedures were performed adhered to the ethical rules and principles of the Helsinki Declaration. The following data were obtained from the hospital's medical records: Data on demographic information, smoking status, comorbidities (hypertension [HT], respiratory disease [chronic obstructive pulmonary disease or asthma], coronary artery disease [CAD],congestive heart failure [CHF], atrial fibrillation [AF], obesity, cancer, renal disease, diabetes mellitus [DM] and, autoimmune- neurodegenerative disease), duration of CS treatment, number of cases followed in ICU, duration of ICU stay and hospitalization, SpO_2 (peripheral oxygen saturation) level, pulse rate, body temperature, number of cases intubated, cases treated with nasal, mask, reservoir mask oxygen and high flow nasal cannula oxygen (HFNCO), immunosuppression (related with chemotherapy, previous CS use, tocilizumab treatment and, sepsis), severity of COVID-19 according to oxygen need, need for oxygen treatment at time of hospitalization and day 1,3 and, 7 and, number of cases died at hospital.

The patients were divided into two groups: those treated with corticosteroids, defined as CS group, and those not, defined as control group. The groups were compared in terms of demographic, clinical, laboratory data and, outcome.

At the first few months of pandemics the treatment with CS was not included in the treatment regimen of COVID-19 patients. The control group was consisted from inpatients hospitalized before the start of CS use for COVID-19 with respiratory distress. The CS group included in-patients treated with CS. The CS treatment was started when the patients suffered from respiratory distress and had a SpO_2 level below 90% in room air at rest/on effort. The dose of CS was, an equivalent dose of0.5-1 mg/kg methylprednisolone/day for 5-7 days in mild cases (who were followed up with nasal oxygen) and 250mg-500mg/day methylprednisolone for 3 days followed with 0.5-1mg/kg/day methylprednisolone which was temporarily decreased and stopped in days/weeks according to the clinical severity. The clinical severity of COVID-19 was determined according to oxygen as follows: mild(room air), moderate (nasal oxygen), severe (reservoir mask oxygen) and, very severe (HFNCO/MV[mechanical ventilation]).

Statistical Analysis

Patient data collected in the study were analyzed with the IBM Statistical Package for the Social Sciences (SPSS) for Windows 23.0 package program (Statistical Package for the Social Sciences, Chicago, IL, USA). Discrete data was given as frequency and percentage. The variables were not normally distributed so the median (range) for continuous data was given as descriptive value. "Mann Whitney U test" was used to compare the two groups. "Pearson Chi-Square Test" was used to compare two categorical groups. "Logistic Regression Analysis" was used to examine the risk factors for mortality and intensive care unit admission. The effect of CS use on survival was examined with Kaplan Meier test and Log Rank test. The results were considered statistically significant when the P value was less than 0.05.

RESULTS

A total of 185 patients (122 CS group and, 63 control group) hospitalized with SARS-CoV-2 pneumonia were included in the study. The comparison of demographic and clinical features was shown in **Table 1**. Patients in the CS group were younger (median age was 63 [30-91] years and 71 [34-91) years, p<0.001).The distribution of both sexes was similar (79 [64.8%] cases in the CS group and 38 [60.3%] cases in the control group were males, p=0.553). The smoking history was less frequent in the CS group (6 [4.9%] cases were current, 7 [5.7%] cases were ex-smoker and 109 [89.3%] cases were none smokers in the CS group while 9 [14.3%] cases were current, 7 [11.1%] cases were ex-smoker and,47 [74.6%]cases were nonsmoker in controls, p=0.028). Among comorbidities obesity was more and DM was less common in CS group (14 [11.5%] vs 0 [0%], and, 34 [27.9%] vs. 29 [46%], p=0.003 and, p=0.013, respectively). The median duration of steroid treatment was 8 (2-31) days. In CS group, the number of cases followed up in ICU and intubated was lower (55 [45.1%] cases vs. 53 [84.1%] cases, p<0.001 and 40 [32.8%] cases vs. 53 [84.1%] cases, p<0.001 respectively), while median time for ICU and hospital stay was longer (13 [1-32] days vs. 6 [1-29] days, p<0.001 and 11 [5-44] days vs.9 [4-35] days, p=0.005 respectively). Cases treated with HFNCO were similar between two groups (30 [24.6%] cases in CS group vs. 23 [36.5%]cases in controls, p=0.127). Immunosuppression due to chemotherapy, previous CS use, tocilizumab treatment and, sepsis were similar between CS and control group (2 [1.6%] cases vs.2 [3.2%] cases, 9 [7.4] cases vs. 2 [3.2%] cases, 18 [14.8%] cases vs. 5 [7.9%] cases, 30 [24.6%] cases vs. 9 [14.3%] cases, p=0.606, p=0.337, p=0.273 and, p=0.150 respectively). Distribution of cases according to oxygen need were similar between CS and control group (Moderate: 78 [63.9%] cases vs. 40 [63.5%] cases, severe: 37 [30.3%] cases vs. 22 [34.9%] cases and, very severe: 7 [5.7]) cases vs. 1 [1.6%] case, p=0.381). The number of cases died were significantly less in CS group (35 [28.7%] cases vs. 53 [84.1%] cases, p<0.001).

Number of cases needed oxygen treatment at time of hospitalization was more in CS group (Nasal oxygen: 60 [49.2%] cases vs.21 [34.4%] cases, mask oxygen16 [13.1%] cases vs.11 [18%] cases, reservoir mask oxygen:21 [17.2%] cases vs.1 [1.6%] case, HFNCO:4 [3.3%] cases vs. 12 [19.7%] cases, MV:1 [0.8%] case vs.1 [1.6%] case respectively, p<0.001).

Table 1. Clinical and demographic features of the two groups

	CS group (N=122)	Control (N=63)	P value
Age (min-max)	63 (30-91)	71 (34-91)	<0.001
Sex			0.553
Male	79 (64.8)	38 (60.3)	
Female	43 (35.2)	25 (39.7)	
Smoking status			0.028
None smoker	109 (89.3)	47 (74.6)	
Smoker	6 (4.9)	9 (14.3)	
Ex-smoker	7 (5.7)	7 (11.1)	
Comorbidities			
HT	66 (54.1)	37 (58.7)	0.548
Respiratory disease	22 (18)	14 (22.2)	0.627
Coronary artery disease	34 (27.9)	17 (27)	0.898
AF	8 (6.6)	5 (7.9)	0.766
Obesity	14 (11.5)	0 (0)	0.003
Cancer	9 (7.4)	5 (7.9)	1
Renal disease	6 (4.9)	8 (12.7)	0.078
DM	34 (27.9)	29 (46)	0.013
Autoimmune-Neurodegenerative disease	7 (5.7)	2 (3.2)	0.720
Duration of steroid treatment (days)	0	8 (2-31)	NA
Number of cases followed in ICU	55 (45.1)	53 (84.1)	<0.001
Time for ICU stay(days)	13 (1-32)	6 (1-29)	<0.001
Time for hospitalization(days)	11 (5-44)	9 (4-35)	0.005
Number of cases intubated	40 (32.8)	53 (84.1)	<0.001
Cases treated with HFNCO	30 (24.6)	23 (36.5)	0.127
Immunosuppression			
Chemotherapy	2 (1.6)	2 (3.2)	0.606
Previous CS use	9 (7.4)	2 (3.2)	0.337
Tocilizumab treatment	18 (14.8)	5 (7.9)	0.273
Sepsis	30 (24.6)	9 (14.3)	0.150
Severity of COVID-19 according to oxygen need			0.381
None	20(16.4)	15(24.6)	
Moderate (nasal)	78 (63.9)	40 (63.5)	
Severe (Reservoir mask)	37 (30.3)	22 (34.9)	
Very severe (HFNCO/MV)	7 (5.7)	1 (1.6)	
Outcome			<0.001
Survival	87 (71.3)	10 (15.9)	
Death	35 (28.7)	53 (84.1)	

* n (%) or median (minimum-maximum). HT: Hypertension, AF: Atrial fibrillation, DM: Diabetes mellitus, ICU: Intensive care unit, HFNCO: High flow nasal cannula oxygen, CS: Corticosteroid, MV: Mechanical ventilation.

On day 1 the median SpO_2 was similar between two groups (88% [72-93] in CS group and, 88% [62-93] in controls, p=0.477). The oxygen need was less in CS group compared to controls (nasal oxygen:55 [45.1%] cases vs. 26 [41.9%] cases, mask oxygen: 18[14.8%] cases vs. 12[19.4%] cases, reservoir mask oxygen: 34 [27.9%] cases vs. 5 [8.1%] cases, HFNCO: 7 [5.7%] cases vs. 2 [3.2%] cases and, MV 2 [1.6] cases vs. 14 [22.6%] cases, p<0.001). The median pulse rate, body temperature, number of leukocytes and, number of lymphocytes were lower in CS group compared to controls (81 [53-135]/min. vs. 88 [59-134]/min. and, 36.5 [35-40]°C vs.36.8 [35.2-39]°C, 7.2x10^3/ml [2.4-24.5] vs. 10.3×10^3/ml [3-55.6], 0.73 [0.13-18.7]×10^3/ml vs. 1[0.1-51.6]×10^3/ml, p=0.047 and p=0.021,p=0.001, p=0.007 respectively). The median serum CRP, IL-6, and ferritin levels were similar between CS group and controls (120.6 [2-350] mg/dl vs.133.5 (3-318] mg/dl, 35.7 [2-297] vs.18 [6-202] and, 729.6 [24.5-2000] vs.356.7 [20.7-36872.5], p =0.602, p=0.518 and, p=0.086 respectively). The median D-Dimer level was lower in CS group (0.6 [0.01-4.8] ng/dl vs.0.9 (0.05-5.7] ng/dl, p=0.028).

On day 3, the median SpO_2 was similar between two groups (93 [70-99%] in CS group and 92 [79-100%] in controls, p=0.875). The need for oxygen support was less in CS group (nasal: 46 [37.7%] cases vs. 9 [14.8%] cases, mask: 9 [7.4%] cases vs. 5 [8.2%] cases, reservoir mask: 22 [18%] cases vs. 5 [8.2%] cases, HFNCO: 19 [15.6%] cases vs. 5 [8.2%] cases and, MV: 10 [8.2%] cases vs. 29 [47.5%] cases, p<0.001). The median pulse rate was similar between two groups, while the body temperature was lower in CS group significantly (81.5/min [50-137] and 36 [35-38]°C in CS group and, 91/min. [43-127] and, 36.5 [35.5-40.3]°C in controls, p=0.099 and, p<0.001 respectively). The median number of leukocytes and lymphocytes were similar (10.36 [2.37-24.14] ×10^3/ml and, 0.65 [0.17-18.34]×10^3/ml in CS group and, 10.08 [1.59-38.6]×10^3/ml and, 0.87 [0.12-14.75]×10^3/ml in controls, p=0.965 and, p=0.098 respectively). The median serum CRP and D-Dimer level was lower in CS group (64.1 [1.6-280.87] vs. 16.6 [2-202], and, 0.57 [0.01-8.43] vs. 0.89 [0.04-4.38], p=0.027 and p=0.039 respectively).The median IL-6 and, ferritin level was similar between two groups (77.7 [2-307] and 706 [87.2-2000] in CS group and, 30.8 [7.71-501] and, 461 [60-100000] in controls, p=0.108 and, p=0.124 respectively).

On day 7 the median SpO_2 value was higher (94 [74-100%] vs. 90 [49-99%], p<0.001) while the median number of cases needed oxygen support was lower in CS group(nasal: 25 [20.5%] cases vs.4 [6.6%] cases, mask: 19 [15.6%] cases vs. 4 [6.6%] cases, reservoir mask: 18 [14.8%] cases vs. 7 [11.5%] cases, HFNCO: 16 [13.1%] cases vs. 2 [3.3%] cases and, MV: 23 [18.9%] cases vs. 41 [67.2%] cases, p<0.001). The median pulse rate was lower in CS group (81 [36.3-140]/min. vs. 91 [36-131]/min., p=0.017) while the median body temperature was similar between two groups (36.4 °C [34.9-39.1°C] in CS group and, 36.5°C [36-39.5°C] in controls, p=0.121).The median number of leukocytes and lymphocytes were also similar (9.69 [2.92-26.25]×10^3/dl and, 0.8 [0.15-18.32] ×10^3/dl in CS group and, 12.1[4.07-213.13]×10^3/dl and, 0.79 [0.1-158.28]×10^3/dl in controls, p=0.202, p=0.873, respectively). The median serum CRP level was lower in CS group (21.48 [0.1-285.69] vs. 89.55 [4.1-326], p<0.001). The median IL-6, D-Dimer and ferritin level were similar (36.90 [2.82-5689], 0.99 [0.01-8.05] ng/ml and, 669 [47.48-2000] in CS group and, 43.3 [5.85-733], 1.44 [0.12-194] ng/ml and, 540.92 [103-1106,9] in controls, p=0.571,p=0.193, p=0.119 respectively) (**Table 2**).

The effect of CS use on survival was examined with Kaplan Meier test and Log Rank test and showed the significant positive effect of CS treatment on survival (**Figure 1**).

Table 2. Oxygen need and laboratory data of two groups

	CS group (N=122)	Control (N=63)	P value
Oxygen need on admission			<.0.001
None	20 (16.4)	15 (24.6)	
Nasal	60 (49.2)	21 (34.4)	
Mask	16 (13.1)	11 (18)	
Reservoir mask	21 (17.2)	1 (1.6)	
HFNCO	4 (3.3)	12 (19.7)	
MV	1 (0.8)	1 (1.6)	
DAY 1			
SpO_2	88 (72-93)	88 (62-93)	0.477
Oxygen need			<.0.001
None	6 (4.9)	3 (4.8)	
Nasal	55 (45.1)	26 (41.9)	
Mask	18 (14.8)	12 (19.4)	
Reservoir	34 (27.9)	5 (8.1)	
HFNCO	7 (5.7)	2 (3.2)	
MV	2 (1.6)	14 (22.6)	
Pulse/minutes	81 (53-135)	88 (59-134)	0.047
Body temperature	36.5 (35-40)	36.8 (35.2-39)	0.021
Number of leukocytes ($\times10^3$/ml)	7.2 (2.4-24.5)	10.3 (3-55.6)	0.001
Number of lymphocytes ($\times10^3$/ml)	0.73 (0.13-18.7)	1 (0.1-51.6)	0.007
CRP (mg/dl)	120.6 (2-350)	133.5 (3-318)	0.602
IL-6	35.7 (2-297)	18 (6-202)	0.518
D-Dimer (ng/ml)	0.6 (0.01-4.8)	0.9 (0.05-5.7)	0.028
Ferritin	729.6 (24.5-2000)	356.7 (20.7-36872.5)	0.086
DAY 3			
SpO_2	93 (70-99)	92 (79-100)	0.875
Oxygen need			<0.001
None	16 (13.1)	8 (13.1)	
Nasal	46 (37.7)	9 (14.8)	
Mask	9 (7.4)	5 (8.2)	
Reservoir mask	22 (18)	5 (8.2)	
HFNCO	19 (15.6)	5 (8.2)	
MV	10 (8.2)	29 (47.5)	
Pulse	81.5 (50-137)	91 (43-127)	0.099
Body temperature	36 (35-38)	36.5 (35.5-40.3)	<0.001
Number of leukocytes ($\times10^3$/ml)	10.36 (2.37-24.14)	10.08 (1.59-38.6)	0.965
Number of lymphocytes ($\times10^3$/ml)	0.65 (0.17-18.34)	0.87 (0.12-14.75)	0.098
CRP(mg/dl)	64.1 (1.6-280.87)	77.7 (2-307)	0.027
IL-6	16.6 (2-202)	30.8 (7.71-501)	0.108
D-Dimer(ng/ml)	0.57 (0.01-8.43)	0.89 (0.04-4.38)	0.039
Ferritin	706 (87.2-2000)	461 (60-100000)	0.124
DAY 7			
SpO_2	94 (74-100)	90 (49-99)	<0.001
Oxygen need			<0.001
None	21 (17.2)	3 (4.9)	
Nasal	25 (20.5)	4 (6.6)	
Mask	19 (15.6)	4 (6.6)	
Reservoir mask	18 (14.8)	7 (11.5)	
HFNCO	16 (13.1)	2 (3.3)	
MV	23 (18.9)	41 (67.2)	
Pulse (/min.)	81 (36.3-140)	91 (36-131)	0.017
Body temperature (°C)	36.4 (34.9-91)	36.5 (36-39.5)	0.121
Number of leukocytes ($\times10^3$/ml)	9.69 (2.92-26.25)	12.1 (4.07-213.13)	0.202
Number of lymphocytes ($\times10^3$/ml)	0.8 (0.15-18.32)	0.79 (0.1-158.28)	0.873
CRP(mg/dl)	21.48 (0.1-285.69)	89.55 (4.1-326)	<0.001
IL-6	36.90 (2.82-5689)	43.3 (5,85-733)	0.571
D-Dimer (ng/ml)	0.99 (0.01-8.05)	1.44 (0,12-19,4)	0.193
Ferritin	669 (47.48-2000)	540.92 (103-1106,9)	0.119

n (%) or median (minimum-maximum). HFNCO: High flow nasal cannula oxygen, MV: Mechanical ventilation, SpO_2: Peripheral oxygen saturation, CRP: C-reactive protein, IL-6: Interleukin 6

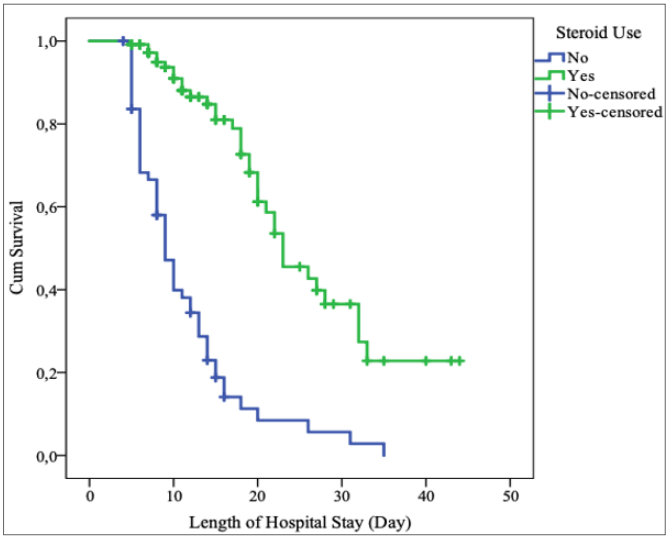

Figure 1. Kaplan–Meier curves of cumulative survival rate for patients administered with and without corticosteroids

DISCUSSION

In this retrospective observational study, the use of CS was related with better prognosis and increased survival in hospitalized patients with COVID-19 pneumonia. Primary endpoint was hospital mortality rate which was significantly lower in CS group although DM and obesity were more frequent in this group. On the other hand CS group was younger than controls. There are several studies reported that both DM, obesity and older age increased the COVID-19 related morbidity and mortality (1,2,13-16). Our findings about positive effect of CS on mortality was compatible with the results of several previous studies (6-10). A meta-analysis including more than 20,000 patients confirmed a beneficial effect of CS treatment on short-term mortality and a reduction in need for MV in hospitalized COVID-19 patients (10). But there are few studies against the use of CS which reported that the CSs did not maintain beneficial clinical outcome and decreased hospital mortality but resulted in adverse effects such as bacterial superinfections and delayed viral clearance (12). As we did not have detailed data on PCR conversion time and complications related to CS treatment we only focused on the effect of the treatment on clinical course and survival rate.

Secondary endpoint was the clinical course. Although the ratio of cases with oxygen need on admission was more in the CS group, on follow up the oxygen need was less and, laboratory parameters related to worse prognosis and mortality were lower in CS group. This may be explained with the potent anti-inflammatory effect of CS.

The number of cases transferred to ICU was more in the control group. CS treatment may prevent the worsening of the cases and decrease the need for ICU follow-up.

Liu et al. (3) reported that although the early start of CS treatment before the need for oxygen support is beneficial, there were no significant differences in hospital mortality rates between patients with and without CS treatment. In our study, we did not furtherly analyze the effect of CS treatment in relation to time of initiation of the treatment but we showed the positive effect of CS on morbidity and mortality.

The duration of hospitalization and ICU follow up was shorter in controls. This may be due to adverse effects of CS such as bacterial superinfections or hyperglycemia which may result in the need for extra treatment and prolonged hospital stay. Also, the ratio of the patients who needed oxygen treatment on admission was more in CS group, so the longer hospital stay may be due to the presence of more severe patients in the study group compared to controls.

There are several studies evaluating the effect of CS treatment on COVID-19 pneumonia, but the results may be confusing and include some bias due to the varying indication, dose and, duration of CS treatment and, different mortality measures (hospital mortality, 28 days mortality, etc.). It was reported that CS might have beneficial effects to reduce the hyper inflammatory response and result in clinical improvement in patients with ARDS (17). On the other hand, the adverse effects, such as delayed viral clearance, opportunistic fungal and bacterial superinfections, and hyperglycemia have to be kept in mind during making the decision to start CS treatment and follow-up. It was reported that CS treatment did not improve survival, reduce hospital stay or ICU admission rate and/or use of mechanical ventilation and prolonged viral clearance (11,12).

This study had a few limitations. The study group was heterogeneous including a large spectrum of patients from mild to very severe cases. The sample size was small so subgroup analysis could not be performed. The patients who received CS were younger which may affect the clinical course and mortality in a positive manner. The adverse effects and complications such as secondary bacterial infections were not evaluated which may be related to clinical outcome and mortality

CONCLUSION

CS treatment is related to a better prognosis and less mortality in inpatients with COVID-19 pneumonia. In addition, it may prolong the duration of hospitalization and ICU follow up. So the decision for CS treatment should be made carefully.

ETHICAL DECLARATIONS

Ethics Committee Approval: This study was approved by Ümraniye Training and Research Hospital Clinical Researches Ethics Committee (Date: 27.05.2021, Decision No: B.10.1.TKH.4.34.H.GP.0.01/179).

Informed Consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

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Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

REFERENCES

1. Hojyo S, Uchida M, Tanaka K, et al. How COVID-19 induces cytokine storm with high mortality. Inflamm Regen 2020; 40: 37.
2. Huang C, Wang Y, Li X, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. Lancet 2020; 395: 497-506.
3. Liu Z, Li X, Fan G, et al. Low-to-moderate dose corticosteroids treatment in hospitalized adults with COVID-19. Clin Microbiol Infect 2021; 27: 112-7
4. Budhathoki P, Shrestha DB, Rawal E, et al. Corticosteroids in COVID-19: is it rational? a systematic review and meta-analysis. SN Compr Clin Med 2020: 1-21.
5. Villar J, Confalonieri M, Pastores SM, et al. Rationale for prolonged corticosteroid treatment in the acute respiratory distress syndrome caused by coronavirus disease 2019. Crit Care Explor 2020; 2: e0111.
6. Alijotas-Reig J, Esteve-Valverde E, Belizna C, et al. Immunomodulatory therapy for the management of severe COVID-19. Beyond the anti-viral therapy: a comprehensive review. Autoimmun Rev 2020; 19: 102569.
7. Jiang S, Liu T, Hu Y, et al. Efficacy and safety of glucocorticoids in the treatment of severe community-acquired pneumonia: a meta-analysis. Medicine (Baltimore) 2019; 98: e16239.
8. Singh AK, Majumdar S, Singh R, et al. Role of corticosteroid in the management of COVID-19: A systemic review and a Clinician's perspective. Diabetes Metab Syndr 2020; 14: 971–8.
9. Veronese N, Demurtas J, Yang L, et al. Use of corticosteroids in coronavirus disease 2019 pneumonia: a systematic review of the literature. Front Med (Lausanne) 2020; 7: 170.
10. van Paassen J, Vos JS, Hoekstra EM, et al. Corticosteroid use in COVID-19 patients: a systematic review and meta-analysis on clinical outcomes. Crit Care 2020; 24: 696.
11. Russell CD, Millar JE, Baillie JK. Clinical evidence does not support corticosteroid treatment for 2019-nCoV lung injury. Lancet 2020; 395: 473–5.
12. Cogliati Dezza F, Oliva A, Cancelli F, et al. Determinants of prolonged viral RNA shedding in hospitalized patients with SARS-CoV-2 infection. Diagn Microbiol Infect Dis 2021; 100: 115347.

13. Wu C, Chen X, Cai Y, et al. Risk factors associated with acute respiratory distress syndrome and death in patients with coronavirus disease 2019 pneumonia in Wuhan, China. JAMA Intern Med 2020; 180: 934-43
14. Şahin Özdemirel T, Akkurt ES, Ertan Ö, Gökler ME, Akıncı Özyürek B. Complications with moderate-to-severe COVID-19 during hospital admissions in patients with pneumonia. J Health Sci Med 2021; 4: 766-771.
15. Güngörer B. Baseline demographic, clinical and laboratory risk factors for predicting admission to intensive care unit in patients diagnosed with COVID-19 in the emergency department. Anatolian Curr Med J 2021; 3; 279-283.
16. Öztürk Durmaz Ş, Sümer Coşkun A, Yalçın AN. Clinical and prognostic evaluation of patients admitted to the COVID-19 pandemic unit of the emergency department. J Health Sci Med 2021; 4: 835-839.
17. Villar J, Ferrando C, Martínez D, et al. Dexamethasone treatment for the acute respiratory distress syndrome: a multicentre, randomised controlled trial. Lancet Respir Med 2020; 8: 267–76.

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The effect of driver age and working hours on increased motorcycle accidents with the COVID-19 pandemic: a cross-sectional case study

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ABSTRACT

Aim: In the present study, the purpose was to investigate the effects of environmental and personal risk factors on accidents in the motor courier business, which expanded with the pandemic.

Material and Method: A total of 227 patients who applied to the Emergency Department after motorcycle accidents between After Pandemic (AP) March 2020-March 2022 and Before Pandemic (BP) March 2018-March 2020 period were included in the study. Statistical differences were analyzed regarding the number of motorcycle accidents before and after the pandemic, the occupation of the patients, driving experiences, weather conditions, and the timing of the accident.

Results: No statistically significant differences were detected between the mean age, gender distribution, occupational distribution, accident occurrence time, and duration of experience of the patients in motorcycle accidents admitted to the Emergency Department ($p>0.05$). Statistically significant differences were detected between the weekly working hours of the patients BP and AP ($p<0.05$). The rate of couriers under the age of 30 and their working time were found to be significantly higher than the other occupational groups in the AP period ($p<0.01$).

Conclusion: The present study showed that the increase in motorcycle accidents with the pandemic did not differ professionally but could be associated with the extended working hours and being under 30 years old. The study can provide useful information on designing accident-avoidance policies and guidelines for motorcycle couriers.

Keywords: Motorcycle couriers, personal risk factors, accident-avoidance policies

INTRODUCTION

A new type of Coronavirus was detected in Wuhan, China in December 2019 with a clinic of atypical pneumonia (1). Because of the rapid spread of this virus, which was called COVID-19, it was later declared a pandemic by the World Health Organization (WHO) (2).

The connection between the service sector and customers was provided by digital systems and couriers in the process after the declaration of the pandemic. Motorcycles are often preferred by couriers because they are less affected by the busy traffic and are easy to park on narrow streets (3). As an important part of the delivery sector in this business line, which is called Motor courier, there has been a significant increase in the demand and the number of employees after the pandemic (4).

Being a motorcycle courier is a physically demanding profession resulting from riding a motorcycle. There are no clear rules defined in terms of daily driving and break times for motorcycle couriers (5). The incomes of the motor couriers are determined according to their working hours and the number of goods they deliver. Couriers can only save their income by making more deliveries because of intense competition. For this reason, the probability of taking risks increases. However, changing personal and environmental factors and unsafe working conditions also increase the risks of accident rates in this regard (6).

The present study aimed to evaluate the hospital emergency department motorcycle accident applications during the pandemic period by comparing the analysis of the patients in terms of occupation, driving experience, weather conditions, and the timing of the accidents during the Before Pandemic period.

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MATERIAL AND METHOD

The study was carried out with the permission of the Marmara University Faculty of Health Sciences, Non-Invasive Clinical Ethics Committee (Date: 24.02.2022, Decision No: 24). The study was carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki. No informed consent form was obtained for since this is a retrospective study. After the first COVID-19 case was detected in our country on March 11, 2020, patients were admitted to the emergency department of our hospital, which is a tertiary trauma center, as in the Before Pandemic period, regardless of COVID-19.

A total of 153 patients who applied to the emergency department In the After Pandemic (AP) period as a result of motorcycle accidents between March 2020 and March 2022, and 74 patients from the same period, were taken as the control group to make the study in the Pre-Pandemic (BP) March 2018-March 2020 reliable, were included in the study. Those who were injured by a motorcycle when on foot, patients who were injured because of alcohol or drug use, and those with incomplete medical records or who did not want to participate in the study were excluded from the study.

Retrospective observational data were obtained from the digital recording system and patient files by an orthopedic assistant and an orthopedic specialist. The demographic data, occupation, driving experience (month), weather (clear/cloudy-snow/rainy), and timing of the accident (day-night) were also recorded.

Statistical differences were analyzed in terms of the number of motorcycle accidents applied to the Emergency Department before and after the pandemic, the occupation of patients, driving experiences, weather conditions, and the timing of the accidents.

Statistical Analyses

The NCSS (Number Cruncher Statistical System) 2020 Statistical Software (Utah, USA) program was used for statistical analysis. When the study data were evaluated, descriptive statistical methods (mean, standard deviation, median, frequency, ratio) and the Shapiro Wilk test and Box Plot Graphics were used in case of compliance of the variables with the normal distribution.

The Mann-Whitney U test was used for intergroup comparisons in non-normally distributed variables. The Pearson Chi-Square test was used to compare the qualitative data. Significance was evaluated at the $p<0.05$ level.

RESULTS

The present study was conducted with a total of 227 cases 7% (n=16) of whom were female and 93% (n=211) male, at Marmara University Training and Research Hospital between 2018 and 2022. The ages of the patients who participated in the study ranged from 14 to 66 with an average of 28.78±10.67 years. The demographic characteristics of the patients are given in **Table 1**.

Table 1. The evaluation of demographic characteristics of the patients

	Group		p
	BP (n=74)	AP (n=153)	
Age			[a]0.498
Mean±sd	29.86±11.78	28.26±10.09	
Median (min-max)	26 (14-64)	25 (16-66)	
Gender			[b]0.220
Female (n=16)	3 (4.1)	13 (8.5)	
Male (n=211)	71 (95.9)	140 (91.5)	
Profession			[b]0.006**
Courier (n=80)	19 (25.7)	61 (25.7)	
For transportation (n=110)	48 (64.9)	62 (40.5)	
Passenger (n=18)	4 (5.4)	14 (9.2)	
Non-courier (n=19)	3 (4.1)	16 (10.5)	

[a]Mann Whitney U Test, [b]Pearson Chi-Square test, **p<0,01

No statistically significant differences were detected between the mean age, gender distribution, and occupation of the patients in motorcycle accidents admitted to the emergency departments in BP and AP period ($p>0.05$).

The evaluation of BP and AP weather conditions, accident times, driver experience times, and weekly working hours are given in **Table 2**. No statistically significant differences were detected between the rates of BP and AP emergency admissions according to the weather conditions ($p>0.05$).

Table 2. The evaluation of BP and AP weather and accident times

	Group		p
	BP	AP	
Weather condition			[b]0.074
Cloudy-Clear (n=157)	57 (77.0)	100 (65.4)	
Rain-Snow (n=70)	17 (23.0)	53 (34.6)	
Accident time			[b]0.485
Daytime (06:00-18:00) (n=143)	49 (66.2)	94 (61.4)	
Night-time (18:00-06:00) (n=84)	25 (33.8)	59 (38.6)	
Experience (months)			[a]0.257
Mean±SD	7.16±13.01	5.80±5.95	
Median (min-max)	3 (0.1-96)	4 (0.2-30)	
Weekly working hours (hrs.)			[a]0.033*
Mean±SD	23.50±22.72	32.96±25.55	
Median (min-max)	13 (2-80)	30 (1-91)	

[a]Mann Whitney U Test, [b]Pearson Chi-Square test, *p<0.05

No statistically significant differences were detected between the time of the accident and the duration of experience of BP and AP patients (p>0.05). Statistically significant differences were detected between the weekly working hours of the patients who applied to the BP and AP emergency department (p<0.05). AP working times were significantly longer (**Figure 1**).

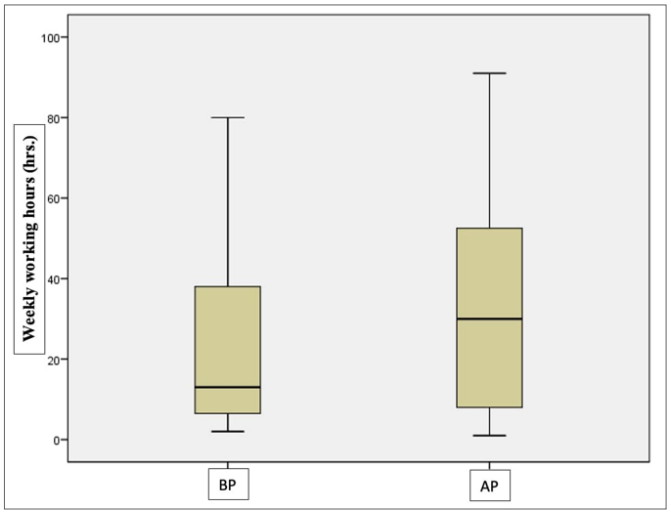

Figure 1. The distribution of BP and AP weekly working hours

The evaluations of the occupations according to age distribution in the BP and AP period are given in **Table 3**. Although no statistically significant differences were detected between age distributions according to occupations in the period of BP (p>0.05); In the AP period, the rate of couriers who were under the age of 30 was found to be significantly higher than other occupational groups (p<0.01).

Table 3. The evaluation of occupations according to age distribution BP and AP

	Age group			P
	<30	30-50	>50	
BP				0.879
Courier	13 (68.4)	4 (21.1)	2 (10.5)	
For transportation	30 (62.5)	13 (27.1)	5 (10.4)	
Passenger	2 (50.0)	2 (50.0)	0 (0)	
Non-courier	3 (100)	0 (0)	0 (0)	
AP				0.008**
Courier	50 (82.0)	9 (14.8)	2 (3.3)	
For transportation	32 (51.6)	26 (41.9)	4 (6.5)	
Passenger	9 (64.3)	3 (21.4)	2 (14.3)	
Non-courier	11 (68.8)	5 (31.3)	0 (0)	

Fisher Freeman Halton test, **p<0,01

DISCUSSION

The first case of COVID-19 was seen in Turkey in the second week of March 2020. After the rapid increase in cases, all countries took measures to protect their citizens and prevent the spread of the disease (7,8). After restaurants closed as part of pandemic measures, our government only allowed motor courier deliveries from restaurants. Also, most supermarkets started motor courier deliveries to reduce social contact and protect against contamination. In the present study, we hypothesized that the increased number of motor courier deliveries manifested itself as a significant increase in the rate of motor courier in motorcycle accidents during the pandemic period. The results of the study were that the motorcycle accident rates in the emergency service admissions increased significant levels by 32.6% in the BP period and 67.4% in the AP period, but the rates of Motor courier in motorcycle accidents did not differ significant levels between the BP and AP periods.

There are many environmental factors affecting traffic accidents of motor couriers (9). The study of Shin et al. (10) on environmental and personal factors in courier accidents showed that 77.2% of courier accidents occurred on cloudy or clear days and 22.8% on rainy or snowy days. A total of 73.5% of the injured couriers had accidents during the day (6:00-18:00) and 26.5% at night (18:00-18:00). In their study, Byun et al. (11) found that the risk of accidents increased in night shifts associated with those working in the courier sector in Korea. In the study of Egozi et al. (4), however, they could not detect significant relations between working conditions and traffic accidents. The difference between the two studies was based on the fact that the couriers in the Korean study were younger, many of them did not have a license, and they mainly worked evening and night shifts. In our study, 69% of motorcycle accidents were on cloudy and clear days, and 31% on rainy and snowy days. Also, 62% of the accidents occurred in the daytime (6:00-18:00) and 38% at night (18:00-18:00). Previous studies are reporting that stress and fatigue factors, which we did not consider in our study, increased the risk of failing to determine the risk in terms of accident and being involved in an accident due to bad decisions (12,13).

Because the barriers to having a job are not difficult for motorcycle couriers, the rate of inexperienced employment without appropriate education and training is high. Shin et al. (10) conducted a study based on the Korean national data bank data in 2019 and showed that 47.2% of the injured couriers had <6 months of work experience and the rate of violations was high (13.9%) among the couriers with <6 months of work experience. Again, Shin et al. (5) In the study, 24.1% of 671 couriers were under the age of 30, 26.5% in their 30s, 24.3% in their 40s, and 25% in their 50s and above. In other words, 75% of them were drivers under the age of 50. In the present study, it was found that although the average work experience was 6 months in the Before Pandemic group, this was around 4 months in the After Pandemic

group. It was also found that 78% of the couriers were drivers under the age of 30, 18.5% were drivers between the ages of 30-50 and 5.7% were drivers over the age of 50. These findings show the need for systematic safety training for inexperienced motorcycle couriers.

A decrease was detected in the total and daily application numbers of trauma patients secondary to the measures taken during the pandemic period (14). Goksoy et al. (15) conducted a study with surgical emergency patients and reported a 25% decrease in admissions. Ilhan et al. (7) showed a 60% reduction in adult trauma patients in their study. Also, Pintado et al. (16) found an 80% decrease in orthopedic trauma applications in their study conducted at a first-level trauma center. Aside from the decrease in general trauma patients in the pandemic period, motorcycle accidents decreased by 39%, as reported in the study of Chiba et al. (17). In the present study, it was found that motorcycle accidents were admitted to the emergency department with a significant increase of 32.6% in the BP period and 67.4% in the AP period. The differences in the studies might have resulted from the trauma patients included in the study from different groups.

The study had some limitations. It had a single-center and retrospective design. Because of the retrospective design, some data were not available. Also, the period selected for the control group was insufficient in terms of comparative data because there were no restrictions on the pandemic. Despite these limitations, the findings show meaningful relations that reinforce the need to explore this industry more deeply to identify areas of professional emphasis and protect safety and health. Further studies are needed to examine the broader scope of risk factors that are associated with the jobs of the couriers beyond the scope of this study.

CONCLUSION

The present study showed that the increase in motorcycle accidents after the pandemic was not occupationally different but could be associated with long working hours and being under 30 years of age, and the working conditions of the couriers were associated with their safety behaviors, safety, and health. The results can provide useful data for designing accident-avoidance policies and guidelines for motorcycle couriers.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of the Marmara University Faculty of Health Sciences, Non-Invasive Clinical Ethics Committee (Date: 24.02.2022, Decision No: 24).

Informed Consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

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Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

REFERENCES

1. Öztürk Durmaz Ş, Sümer Coşkun A, Yalçın AN. Clinical and prognostic evaluation of patients admitted to the COVID-19 pandemic unit of the emergency department. J Health Sci Med 2021; 4: 835-9.
2. Cucinotta D, Vanelli M. WHO Declares COVID-19 a Pandemic. Acta bio-medica Atenei Parmensis 2020; 91: 157-60.
3. Chung Y, Song TJ, Yoon BJ. Injury severity in delivery-motorcycle to vehicle crashes in the Seoul metropolitan area. Accident Analysis & Prevention 2014; 62: 79-86.
4. Egozi L, Reiss-Hevlin N, Dallasheh R, Pardo A. Couriers' safety and health risks before and during the COVID-19 pandemic. Int Arch Occup Environ Health 2022; 95: 589-98.
5. Shin DS, Park MH, Jeong BY. Human factors and severity of injury of delivery truck crashes registered for work-related injuries in South Korea. Industrial Engineering and Management Systems 2018; 17: 302-10.
6. Byun JH, Yong BY, Park MH. Characteristics of Motorcycle Crashes of Food Delivery Workers. J Ergonomics Soc Korea 2017; 36: 157-68.
7. İlhan B, Bozdereli Berikol G, Aydın H, Arslan Erduhan M, Doğan H. COVID-19 outbreak impact on emergency trauma visits and trauma surgery in a level 3 trauma center. Ir J Med Sci 2021; 7: 1–6.
8. Güngörer B. Baseline demographic, clinical and laboratory risk factors for predicting admission to intensive care unit in patients diagnosed with COVID-19 in the emergency department. Anatolian Curr Med J 2021; 3: 279-283.
9. Clarke DD, Ward P, Bartle C, Truman W. The role of motorcyclist and other driver behaviour in two types of serious accident in the UK. Accident Analysis Prevention 2007; 39: 974-81.
10. Shin DS, Byun JH, Jeong BY. Crashes and traffic signal violations caused by commercial motorcycle couriers. Safety and Health at Work 2019; 10: 213–8.
11. Byun JH, Park MH, Jeong BY. Effects of age and violations on occupational accidents among motorcyclists performing food delivery. Work (Reading, Mass.) 2020; 65: 53–61.
12. Day AJ, Brasher K, Bridger RS. Accident proneness revisited: the role of psychological stress and cognitive failure. Accident Analysis Prevention 2012; 49: 532-5.
13. Smith TD, Hughes K, Dejoy DM, Dyal MA. Assessment of relationships between work stress, work-family conflict, burnout and firefighter safety behavior outcomes. Safety Sci 2018; 103: 287-92.
14. İlhan B, Bozdereli Berikol G, Dogan H. Impact of COVID-19 outbreak on emergency visits and emergency consultations: a cross-sectional study. Cureus 2021; 13: e14052.

15. Göksoy B, Akça MT, Inanç ÖF. The impacts of the COVID-19 outbreak on emergency department visits of surgical patients. COVID-19 salgınının cerrahi hastaların acil servis başvuruları üzerine etkisi. Ulus Travma Acil Cerrahi Derg 2020; 26: 685-92.
16. Pintado JF, Gibaja W, Vallejos RA, Rosas W, Guerra Farfan E, Nunez JH. How COVID-19 has affected emergent visits to a Latin-American trauma department: experience at a Peruvian national trauma referral center. Injury 2020; 51: 2834-9.
17. Chiba H, Lewis M, Benjamin ER, et al. Safer at home: the effect of the COVID-19 lockdown on epidemiology, resource utilization, and outcomes at a large urban trauma center. J Trauma Acute Care Surg 2021; 90: 708-13.

Original Article

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The effect of malignancy on prognosis in ICU patients with COVID-19

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ABSTRACT

Background: It is known that COVID-19 has a worse and poorer progression, which often might lead to death in those with comorbidities. Multiple studies have recently examined the clinical course of cancer patients with COVID-19 and new guidelines have been developed to manage this group of patients. This study aimed to evaluate the clinical course and mortality rate of cancer patients admitted to the intensive care unit (ICU) for COVID-19.

Material and Method: The demographic characteristics, detailed medical history and laboratory findings of 140 patients with malignancy, who were treated in the COVID-19 Intensive Care Unit of Ankara City Hospital, were evaluated retrospectively. Gender, age, comorbidity, length of stay in the ICU, mortality rates, length of stay on a mechanical ventilator, cytokine storm scores, ferritin, interleukin 6 (IL-6), C-reactive protein (CRP), procalcitonin (PCT), D-dimer, lactate dehydrogenase (LDH), lymphocyte count and treatment options were compared. The patients were divided into two groups: solid and hematological malignancies.

Results: One hundred eight of 140 patients were diagnosed with solid organ malignancy and 32 with hematological malignancy. The most common comorbidity was found to be hypertension. A total of 94 patients died during their ICU stay. While the most common solid organ malignancies were malignancies of the lower gastrointestinal tract and lung cancers, multiple myeloma (MM) was the most common hematological malignancy. There was no significant difference between the two groups in terms of cytokine storm scores , duration of hospitalization and mechanical ventilation. Levels of Ferritin and LDH were found to be significantly higher in patients with hematological malignancies, while D-dimer was significantly higher in solid organ malignancies. A high level of CRP and IL-6 was associated with COVID-19 mortality. Lymphopenia was associated with increased mortality in patients with solid organ malignancy. However, there was no difference in mortality rate among both groups. Although the mortality was significantly higher in the patient group receiving chemotherapy, there were no significant differences in mortality for the duration of receiving chemotherapy.

Conclusion: The results of this study suggested that cancer was associated with severe clinical outcomes and a 67% mortality rate among patients with COVID-19. However, despite the changes in patients' demographic, clinical and laboratory characteristics, no difference in mortality rate was detected in patients with hematological and solid organ malignancies due to COVID-19 infection.

Keywords: COVID-19, coronaviruses, hematological malignancies, malignancy, solid malignancies, mortality

INTRODUCTION

Coronavirus disease 2019 (COVID-19) disease started in the Wuhan region of China in December 2019, spread all over the world and was declared a pandemic by the World Health Organization (WHO) as of March 12, 2020 (1-3). COVID-19 can be asymptomatic depending on the host's immune response and comorbidities or it can cause life-threatening multi-organ dysfunction. COVID-19 can affect all body areas (especially the respiratory system, arterial-venous system endothelial cells, lymphoid tissue, glial cells in the brain, heart, bone, spleen, urinary tract, intestines, nasal mucosa, skin and muscle tissue). Fever, cough and shortness of breath, which can progress to organ failure are the most common symptoms of COVID-19. In respiratory failure, which occurs with clinical worsening in the later stages of the disease, follow-up in the intensive care unit (ICU) is required according to the need for respiratory support (4). The mortality rate is high in patients with COVID-19. Advanced age, male sex, hypertension, immunodeficiency due to chronic diseases and a history of cancer are the factors affecting mortality (5).

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The COVID-19 pandemic has affected the publications in the literature (6,7). Studies investigating the etiopathogenesis and prognostic factors for COVID-19 in those with underlying medical conditions increase rapidly. Comorbidity is identified as an independent poor prognostic factor for patients with COVID-19. COVID-19 uses ACE-2 receptors on the surface of host cells to enter the cell and there are studies in the literature indicating that certain comorbidities are associated with a strong ACE-2 receptor expression and higher release of proprotein convertase release (8,9). It is reported that COVID-19 patients with comorbidities have worse lung damage and more mortality rate and the most common comorbidities are hypertension, coronary artery disease and diabetes mellitus (9).

In a study in which comorbidities were evaluated, it was determined that 58.3% of COVID-19 patients had lung cancer and 41.7% of them received immunotherapy, chemotherapy or radiotherapy (8). Patients with malignancies were considered to be at higher risk for COVID-19 infection because of their immune deficiency.

The mortality rate in COVID-19 patients diagnosed with malignancy was calculated as 5.6% and it was found to be 3.5 times higher when compared to the general population (10). The mortality rate was found to be 37% in COVID-19 patients with hematological malignancies (11). It has been stated that hematological malignancies are in the higher risk group for COVID-19 due to age, immune system deficiency and risk of other infections (12). Since acute myeloid leukemia (AML) patients under therapy are highly immunosuppressive and their treatment takes a long time, they are at an incredibly high risk of developing life-threatening complications if infected with COVID-19 (13).

Studies show that patients with cancer appear to be at increased risk of mortality and severe illness due to COVID-19 infection. This study aims to provide information about the effects of COVID-19 disease in malignancy patients and to evaluate the clinical differences between hematological and solid organ malignancies.

MATERIAL AND METHOD

The study was carried out with the permission of Ankara City Hospital No 1 Clinical Researches Ethics Committee (Date: 15.12.2021, Decision No: E1-21-2227). All procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki.

Participants

All patients older than 18 were diagnosed with solid or hematologic malignancies prior to admission to the ICU with the diagnosis of COVID-19 who received treatment in the COVID-19 Intensive Care Unit of Ankara City Hospital between 01.03.2020 and 01.01.2022 were included in the study. Participants were divided into two groups, hematological and solid organ malignancies and their data were analyzed retrospectively.

Study Design

Clinical follow-up data were recorded by retrospective case-note review. Firstly, COVID-19 patients with malignancies were evaluated according to their general demographic and clinical and laboratory findings. Then, they were divided into two groups, hematological and solid organ malignancies. Variables such as gender, age, comorbidity, length of stay in the ICU, mortality rates, length of stay on a mechanical ventilator, cytokine storm scores, Ferritin, IL-6, CRP, PCT, D-dimer, LDH, lymphocyte count and treatment were compared.

Statistical Analysis

SPSS (Statistical Package for Social Sciences) for Windows 16.0 program was used for statistical analysis. The compatibility of the parameters with the normal distribution was evaluated with the Kolmogorov-Smirnov test. Descriptive statistics included frequency (n), mean, standard deviation, minimum, median and maximum values. Frequencies and percentages were given for categorical variables. Kruskal-Wallis test was used for comparisons of parameters that did not show the normal distribution in quantitative data. Mann-Whitney U test was used for comparisons between the two groups. Independent-Samples T-test was used for comparisons between two groups with normal distribution. Chi-square test analysis was used to compare the relationship between qualitative data. The results were evaluated at the 95% confidence interval and the significance was at the $p<0.05$ level.

RESULTS

One hundred forty patients with malignancy, who were treated in the COVID-19 Intensive Care Unit of Ankara City Hospital because of COVID-19 pneumonia were evaluated retrospectively (**Figure**). The demographic and clinical characteristics are presented in **Table 1**. One hundred eight of 140 (77.2%) patients were diagnosed with solid organ malignancy and 32 (22.8%) with hematological malignancy. The mean age was 70.2±12.4 (37-96 years). The most common comorbidity was found to be hypertension in 66 (47.1%) patients. Although 88 (62.8%) of the patients were given high flow nasal oxygen therapy, 94 (67%) of them needed mechanical ventilation. Ninety-four (67%) patients died during their stay in ICU.

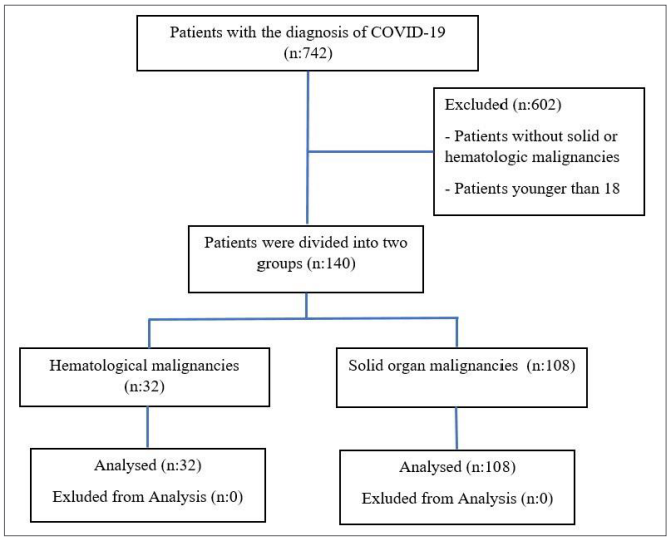

Figure. Flowchart of the patients

Table 1. The demographic and clinical characteristics

Variables	All patients (n=140)
Age	70.2±12.4 (37-96 years)
Gender (Male/female)	80 /60
Hematological malignancy	32 (22.8%)
Mean APACHE score	11.5±6.2
Mean SOFA score	5.8±3.5
Cytokine storm scores (SFS)	3.5±1.2
Duration of hospital stay	10.7±8.1 (1-40 days)
Duration of intubation	4.7±6.8 (0-35 days)
Percentage of intubation	94 (67%)
Percentage of exitus	94 (67%)
Diyabetes mellitus	38 (27.1%)
Hypertansion	66 (47.1%)
Coronery artery disease	37 (26.4%)
Chronic obstructive pulmonary disease	14 (10%)
Chronic kidney failure	9 (6.4%)
Neurological disorder	25 (17.8%)
High flow oxygen therapy	88 (62.8%)
Non-invasive mechanical ventilation	22 (15.7%)
Ferritin	1473±3157
C-reaktive protein	130±89
IL-6	267±1027
D-dimer	5.2±8.7
LDH	599±558
The lymphocytes count	2.9±13.5
Procalsitonin	2.9±11.2

The patients were divided into two groups: solid and hematological malignancies. Staging could not be performed because imaging methods (positron emission tomography -computerized tomography scans) were not available in the vast majority of patients with hematological malignancies. Solid organ malignancies were divided into subgroups among themselves; however, staging was difficult due to the same reason, so they were divided into metastatic and non-metastatic cancer groups. Sixty (55.5%) of 108 patients were found to have metastatic cancers. The most common malignancies were lower gastrointestinal tract (20 patients, 18.5%) and lung cancers (20 patients, 18.5%). In the haematological malignancy group, nine patients (28.1%) had MM, seven patients (21.8%) had lymphoma, six patients (18.7%) had chronic lymphocytic leukaemia (CLL), three patients (9.3%) had chronic myocytic leukaemia (CML), three patients (9.3%) had AML, two patients (6.2%) had myelofibrosis, one patient (3.1%) had acute lymphocytic leukaemia (ALL) and one patient (3.1%) had myelodysplastic syndrome (MDS). All CML patients were discharged, while AML, myelofibrosis, ALL and MDS patients died in the hospital. The mortality rate was 55% in MM patients, 83% in CLL patients and 71%in lymphoma patients. **Table 2** shows the solid malignancy subgroups and and existence of metastasis.

Table 2. The solid malignancy subgroups and the existence of metastasis.

Solid organ malignancies (n=108)	Metastatic (n=60) (55.5%)	Non-metastatic (n=48) (44.5%)	Number of exitus
Gastrointestinal malignancy (n=20)	8	12	13 (11.9%)
Lung cancer (n=20)	14	6	15 (13.8%)
Breast cancer (n=18)	10	8	9 (8.3%)
Prostat cancer (n=13)	5	8	11 (10.1%)
Stomach cancer (n=8)	7	1	6 (5.5%)
Bladder cancer (n=6)	3	3	5 (4.6%)
Pancreas cancer (n=4)	3	1	4 (3.7%)
Intracranial tumours (n=4)	2	2	2 (1.8%)
Renal malignancy (n=3)	1	2	1 (0.9%)
Larynx cancer (n=2)	0	2	1 (0.9%)
Liver cancer (n=2)	2	0	0 (0%)

Ninety-three patients (66.4%) did not receive active chemotherapy (CT) (>1 year), 20 patients (14.3%) had CT within 15 days, 11 patients (7.9%) received CT within 15 days-1 month and 16 patients (11.4%) had been on CT for more than 1 month. (However, there were cured malignancies as well as end-stage malignancies in the group that did not receive CT). When the patients were divided into two groups as those who received CT and those who did not, the mortality rate was found to be significantly higher in the group of patients undergoing chemotherapy (p=0.03). However, no significant correlation was found between the time of the last CT and the mortality rate (p=0.12).

When the patients were compared in terms of hematological and solid organ malignancies, the mean age of patients with hematological malignancies was 65.6±14.4 years and they were statistically younger than the patient group with solid malignancy (p=0.02). The male gender predominated in both groups. The APACHE score was significantly lower in patients with hematological malignancies at 10.2±6.8 (p=0.02). There was no significant difference between SOFA score and cytokine storm score in both groups (p=0.71, p=0.42). There was no significant

difference between the duration of hospitalization and mechanical ventilation, nor in terms of mortality rate (p=0.31, p=0.93, p=0.93).

Elevated serum ferritin and LDH levels have been found in significantly higher in hematological malignancies (p=0.04 and p=0.04). D-dimer was significantly higher in solid organ malignancies (p=0.01) (**Table 3**). Levels of PCT, IL-6 and aspartate amino transferase (AST) in hematological malignancies were significantly higher in patients who died (p=0.01, p=0.01, p=0.04, retrospectively). In solid organ malignancies, PCT and IL-6 values were found to be significantly higher in patients who died in the ICU (p=0.009 and p=0.007), while the lymphocyte count was significantly lower in this patient group (p=0.03).

Additionally, Intravenous immune globulin (IVIG) therapy was used in 16.1% of patients with hematological malignancies, while 1.8% of patients with solid organ malignancies had received this treatment (p=0.006).

DISCUSSION

Research on the course of Covid-19 disease in special disease groups such as cancer patients is limited. It is known that the high mortality rate in COVID-19 patients is associated with their comorbid conditions (9). The clinical course of the COVID-19 in malignancy patients, which is a particular group, was evaluated in our study, especially the clinical outcome between solid organ and hematological malignancies. The mortality rate was found to be significantly higher in cancer patients receiving CT. No significant difference was detected.

A study published in JAMA stated that 2,523,920 patients out of 73.4 million patients were diagnosed with malignancy. Of these patients, 16,570 were diagnosed with COVID-19 disease and 1200 patients had malignancy. In patients diagnosed within <1 year, especially leukemia, non-Hodgkin lymphoma and lung cancers were found to be associated with increased COVID-19 disease risk (p<0.01). In addition, it has been shown that patients with malignancy and COVID-19 have a higher duration of hospital stay and mortality rates (p<0.01, p<0.01) (13).

A study by Mehta et al. (11) reported that 61 patients (28%) of 218 cancer patients diagnosed with COVID-19 died in their ICU stay. Among them, the mortality rate was 37% (20/54) in those with hematological malignancies, while it was 25% in those with solid organ malignancies. They found that the mortality rate increased significantly when compared with patients without malignancy. In the same study, although the mortality rate was higher in lung and gastrointestinal cancers, it was reported that the mortality rate was lower in genitourinary and breast cancers. Among hematological malignancies, myeloid-derived hematological cancers were found to be a higher mortality rate than lymphoid-derived malignancies. The numbers of intensive care unit admission and ventilator use were slightly higher for hematological malignancies than for solid tumors (26% vs 19% and 11% vs 10%, respectively), but this finding did not show a statistical significance (11). While it was expected that patients with metastasis and newly diagnosed patients (<1 year) would show an increased mortality rate, no statistical

Table 3. Comparison of demographic, clinical and laboratory data of patients with hematological malignancies and solid organ malignancies

	Hematological malignancies (n=32)	Solid organ malignancies (n=108)	P value
Age	65.6±14.4	71.4±11.6	<0.02
Gender (Male)(n=80)	16 (20%)	64 (80%)	>0.54
APACHE score	10.2±6.8	11.8±6.0	<0.02
SOFA score	55±31	58±36	0.71
Cytokine storm scores	36±10	34±13	0.42
Duration of hospital stay (days)	92±59	112±85	0.31
Duration of intubation (days)	40±56	49±71	0.93
Percentage of intubation (n=94)	22 (68.7%)	72 (66.6%)	0.83
Percentage of exitus (n=94)	22 (68.7%)	72 (66.6%)	0,93
Diyabetes mellitus (n=38)	8 (21%)	30 (79%)	1.0
Hypertansion (n=66)	12 (18.1%)	54(71%)	0.31
Coronery artery disease (n=37)	4 (10%)	33 (90%)	0.06
Chronic obstructive pulmonary disease (n=14)	2 (14.2%)	12 (85.8%)	0.73
Chronic kidney failure (n=9)	1 (11.1%)	8 (88.9%)	0.68
Neurological disorder (n=25)	2 (8%)	23 (92%)	0.06
Ferritin	3014±5962	1035±1442	0.04
C-reaktive protein	1377±942	1285±888	0.56
IL-6	423±1559	2231±812	0.18
D-dimer	22±18	61±97	0.01
LDH	704±802	569±468	0.04
The lymphocytes count	83±256	13±64	0.62
Procalsitonin	57±205	21±63	0.82

significance was found (p=0.06 and p=0.09) (11). The current study calculated the total mortality rate as 67%. This rate was 68.7% in patients with hematological malignancies and 66.6% (72 out of 108) in solid organ malignancies. However, there was no statistical significance. The most common malignancies were lower gastrointestinal and lung cancers and mortality rates were higher in correlation with this study. There was no difference between the two groups regarding the duration of the ICU stay and intubation days. In parallel with Mehta et al. (11) there was no significant difference in the mortality rates of patients with metastatic disease compared to those with non-metastasis.

In a case series in which 67 patients with malignancy were evaluated, 23 patients had received active chemotherapy. The most common type of cancer was lung cancer (22.4%). It was reported that the clinical outcomes of COVID-19 disease were more severe in elderly patients and it was determined that these patients had several comorbidities. In addition, the authors stated that patients receiving active CT had a worse prognosis (14).

In a study including 28 cancer patients by Zhang et al. (15) the authors reported that patients who received CT in the last 14 days had a worse prognosis. The mortality rate was 26% and patients requiring invasive mechanical ventilation were 36%. The most common malignancy was lung cancer, which was correlated with other studies. It was also stated that 71% of these patients were given IVIG therapy (15). In our study, the intubation rate was 67% and the mortality rate was 67%. Our results showed that 66.4% of patients did not receive active CT (>1 year) and 33.6% received CT within the last year. While mortality rates were higher in patients receiving CT, no significant correlation was found with the time of receiving CT. Although the mean age and APACHE-II scores were significantly lower in hematological malignancies, there was no significant difference in mortality rate compared to solid organ malignancies. IVIG was given to 17.9% of patients and it was most frequently applied to patients with hematological malignancies.

In June 2021, the data of 1013 patients infected with COVID-19 were shared by the American Society of Hematology (ASH) and it was stated that the mortality rate ranged between 13.8% and 39% (16). Various clinical and laboratory parameters such as one or more comorbidities, age, type of malignancy (especially AML), high level of CRP, lymphopenia and neutropenia have also been reported to be risk factors for mortality in these patients (17). The most common subgroup of hematological malignancy was MM in our study. The most common mortality rate was seen in CLL patients (83%). However, there were 2 AML patients and both of them died in the ICU. When the laboratory data were analyzed, it was found that mortality was higher in patients with elevated levels of PCT, IL-6 and AST. No significant correlation was found between lymphocyte count and mortality. In addition, patients with solid organ malignancies had higher procalcitonin and IL-6 values and severe lymphopenia. While these results are in agreement with the literature data, no significant relationship was found between lymphocyte count and mortality rate.

In a study that included 3801 patients with COVID-19 and hematological malignancies with data from 132 centers, the most common subgroup was found to be non-Hodgkin Lymphoma (1084, 28.5%) and the second most common malignancy was MM (684, 18%). 73.1% of these patients were hospitalized. The average hospital stay was 15 days. 18.1% of patients needed intensive care and 65.2% were intubated. The number of patients whose primary cause of mortality was COVID-19 pneumonia was evaluated as 155 (13.1%) (18). Although the duration of stay in the ICU was not shared in this study, it was 9.2±5.9 days in our study. We found a 68.7% intubation and mortality rate. In the current study, we found that all patients with solid organ malignancies and hematological malignancies who were intubated for mechanical ventilation died during their stay in ICU. When hematological and solid organ malignancies were compared, there was no difference in mortality rate. However, when compared with this study, it can be argued that the mortality rate was higher since primary causes of mortality were not distinguished in our study. In addition, since IL-6 and PCT levels had a significant effect on mortality in both groups, it might be thought that sepsis often accompanies these patients. We detected that severe lymphopenia was significantly related to mortality in patients with solid organ malignancies. However, the evaluation of lymphopenia in hematological malignancies is complex and careful consideration should be given to using it as a basis for treatment and prognostic evaluation.

CONCLUSION

In conclusion, the mortality rate of COVID-19 patients with malignancy is high. No significant difference was found between hematological malignancies and solid organ malignancies in terms of mortality in our study. Since the mortality rate was high in patients undergoing chemotherapy, which was similar to other studies' findings, it can be recommended that primary prevention methods related to COVID-19 and special attention must therefore be given among this group. Randomized and prospective trials, including homogeneous patient groups, are needed to evaluate the data more accurately.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Ankara City Hospital No 1 Clinical Researches Ethics Committee (Date: 15.12.2021, Decision No: E1-21-2227).

Informed Consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

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Author Contributions: All of the authors declare that they have all participated in the design, execution and analysis of the paper and that they have approved the final versio.

REFERENCES

1. Gürsoy C, Oral Tapan Ö, Doğan E, Pektaş S, Demirbilek S. Early and late results of intravenous immunoglobulin as potential adjuvant therapies in critically ill COVID-19 patients: a retrospective cohort study. J Health Sci Med 2022; 5: 794-8.
2. Gemcioğlu E, Başer S, İnan O, et al. Effects of blood group types on risk of infection, disease severity and mortality in COVID-19 patients. J Health Sci Med 2022; 5: 374-8.
3. Ayyıldız A, Çobaner N, Erben N, Yelken B. A retrospective, observational study: early versus late favipiravir in COVID-19 pneumonia. J Med Palliat Care 2022; 3: 22-5.
4. Mao L, Jin H, Wang M, et al. Neurologic manifestations of hospitalized patients with coronavirus disease 2019 in Wuhan, China JAMA Neurol 2020; 77: 683-90.
5. Zhou F, Yu T, Du R, et al. Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study. Lancet 2020; 395: 1054-62.
6. Zengin M, Karcioglu AM. Do not invade, just support. Bratisl Lek Listy 2022; 123: 218-26.
7. Zengin M, Baldemir R. Investigation of the global outcomes of acute respiratory distress syndrome with the effect of COVID-19 in publications: a bibliometric analysis BETWEEN 1980 and 2020. KırıkkaleÜniversitesi Tıp Fakültesi Derg 2021; 23: 279-92.
8. Ejaz H, Alsrhani A, Zafar A et al. COVID-19 and comorbidities: Deleterious impact on infected patients. J Infect Public Health 2020; 13: 1833-9.
9. Zhou F, Yu T, Du R, Fan G, Liu Y, Liu Z. Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study. Lancet 2020; 395: 1054-62.
10. Liang W, Guan W, Chen R, Wang W, Li J, Xu K. Cancer patients in SARS-CoV-2 infection: a nationwide analysis in China. Lancet Oncology 2020; 21: 335–7.
11. Mehta V, Goel S, Kabarriti R, et al. Case fatality rate of cancer patients with COVID-19 in a New York Hospital System. Cancer Discov 2020; 10: 935-41
12. Zeidan AM, Boddu PC, Patnaik MM, et al. Special considerations in the management of adult patients with acute leukaemias and myeloid neoplasms in the COVID-19 era: recommendations from a panel of in-ternational experts. Lancet Haematology 2020; 7: e601-12.
13. Wang Q, Berger NA, Xu R. Analyses of Risk, racial disparity and outcomes among US patients with cancer and COVID-19 infection. JAMA Oncology 2021; 7: 220–7.
14. Zhang H-Y, Wang L-W, Chen Y-Y, et al. A multicentre study of 2019 novel coronavirus dise-ase outcomes of cancer patients in Wuhan, China. medRxiv (2020) (Epub ahead of print).
15. Zhang L, Zhu F, Xie L, et al. Clinical characteristics of COVID-19-infected cancer patients: a retrospective case study in three hospitals within Wuhan, China. Ann Oncol2020; 31: 894-901.
16. https://www.ashresearchcollaborative.org/s/covid-19-registry/data-summaries(Last access: May 25, 2021)
17. García-Suárez J, De la Cruz J, Cedillo Á, et al. Impact of hematologic malignancy and type of cancer therapy on COVID-19 severity and mortality: lessons from a large population-based re-gistry study. J Hematol Oncol 2020; 13: 133.
18. Pagano L, Salmanton-García J, Marchesi F, et al. COVID-19 infection in adult patients with hematological malignancies. J Hematol Oncol 2021 14; 14: 168.

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Evaluation of survival outcomes and prognostic factors in acinic cell carcinomas of the parotid gland receiving adjuvant radiotherapy

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ABSTRACT

Aim: To evaluate the survival outcomes and prognostic factors in acinic cell carcinoma of the parotid gland, a retrospective study was designed.

Material and Method: Consecutive patients diagnosed with parotid acinic cell carcinoma and treated with surgery and adjuvant radiotherapy were retrospectively reviewed. Data regarding age, sex, TNM stage, pathologic characteristics, treatment details, and follow-up examinations were collected and analysed. The primary end-point was overall survival; the distant metastasis free survival was calculated from the date of surgery to the date of death or the latest follow-up examination and analysed by the Kaplan-Meier method. Independent prognostic factors were evaluated by the Cox proportional hazards method.

Results: Between years of 2010-2020, two radiotherapy centers' database were reviewed. A total of 32 patients were included. The median age was 55 years (35-80 years). Four-teen (43.75%) were male and 18 (56.25%) were female. Median follow-up was 44 months (8-120). Seven (21.9%) were in T1, 7 (21.9%) in T2, 6 (18.8%) in T3 and 12 (37.5%) in T4 at the time of diagnosis. In all cohort, 6 (18.8%) of them had lymph node metastasis. The 2-year and 5-year OS rates were 92.6% and 78.5%, locoregional recurrence-free survival rates were 100% and 89.1%, distant metastasis free survival rates were 85.9% and 85.9%, respectively. Locoregional recurrence detected in 2 (6.25%), distant metastases detected in 4 (12.5%) patients. All distant metastases detected in the lungs. Univariate analysis showed that age, gender, margin status, T stage, facial nerve involvement, lymphovascular invasion, and perineural invasion were not significantly related to overall survival (all $p>0.05$). Lymph node involvement ($p<0.013$) and grade ($p<0.006$) were the only significant prognostic factors for OS. In multivariate analysis, both lymph node involvement ($p<0.050$) and grade ($p<0.028$) remained the significant prognostic factors.

Conclusion: In acinic cell carcinoma of the parotid gland, high-grade histology and node positivity are independent variables that affect OS. Since survival is lower in these patient groups, it is imperative to explore other treatment options in addition to adjuvant radiotherapy.

Keywords: Parotid cancer, acinic cell carcinoma, prognostic factors, radiotherapy, survival

INTRODUCTION

Acinic cell carcinoma (ACCs) is a very rare malignant tumor. The vast majority are located in the parotid gland (86% of cases) (1) and constitute 6-16% of all malignant tumors originating from the major salivary glands (2). It was defined as benign adenoma by Goodwin et al. (3) in 1890, recognized as a malignant tumor by Buxton et al. (4) in 1953 and is currently classified as malignant by World Health Organization (5). Although it's been considered for a long period of time a neoplasm with a good prognosis, currently, we should take into account it as a malignancy with an uncertain clinical course, since this tumor tends to recur, metastasize, and even cause death, particularly for a subgroup, defined as "ACCs with high-grade transformation".

The prognostic factors in ACCs are not well documented, due to the small sample size, poor quality of medical records, and difficulty in organizing randomized trials. In this study, we retrospectively evaluated survival outcomes and prognostic factors in parotid ACCs who received surgery and adjuvant radiotherapy (RT). Medical records of two RT centers were collected for increasing the sample size.

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MATERIAL AND METHOD

The study was conducted in accordance with the ethical principles of the Declaration of Helsinki and was approved by a local human research committee. The study protocol was reviewed and approved by the Clinical Research Ethics Committee (Date: 12/04/2022, Decision No: 2022/176). Written informed consent forms were read by each patient and signed consent was obtained prior to their treatment. Medical records of two RT centers were reviewed. Between January 2010 and January 2020, a total of 32 cases with ACCs, who met with the inclusion criteria, included in the study.

Inclusion and Exclusion Criteria

Inclusion criteria:

- Who diagnosed with ACCs in parotid gland,
- Patients were> 18 years of age
- Who underwent a currative surgery,
- Who received adjuvant RT,
- Cases without a postoperative macroscopic residual mass,
- Cases who have not received neoadjuvant, adjuvant or concurrent chemotherapy

Exclusion criteria:

- Relapsed disease prior to adjuvant RT,
- Cases with no surgery for curative intent,
- Cases with a previous history of another malignant disease,
- Who developed a second primary malignancy during followup period,
- Cases with metastases prior to RT,
- Cases with postoperative macroscopic residual mass (R2 resection),
- Cases with immunosuppressive disease.

Statistical Analysis

Study data were analyzed using the statistical package program Statistical Package for the Social Sciences version 25.1 (SPSS, Inc., Chicago, IL, ABD). Numeric, percentage, standard deviation, mean, minimum and maximum values were used as descriptive statistics. Locoregional recurrence-free survival (LRFS), distant metastasis-free survival (DMFS), and overall survival (OS) were estimated using the Kaplan-Meier method. To identify prognostic factors that might affect survival, log rank tests were performed to examine univariate relationships between survival and parameters of interest, and Cox regression analysis to examine multivariate relationships. A value of $p<0.05$ was considered statistically significant.

RESULTS

Patient and Histopathological Characteristics and Treatment Outcomes

We analyzed overall outcomes and investigated potential prognostic variables such as age, gender, T stage and N stage, facial weakness, histologic grade, extraglandular spread, resection margins, perineural invasion and lymphovascular invasion. The date of diagnosis was accepted as the histological diagnosis date. The last follow-up date was the last consultation date. All tumors were classified by the tumor (T), lymph node (N), and metastasis staging system, seventh edition (International Union Against Cancer, 2009). Of 32 patients, all were located in parotid glands. The median age was 55 years (35-80 years). Four-teen (43.75%) were male and 18 (56.25%) were female. Median follow-up was 44 months (8-120). Seven (21.9%) were in T1, 7 (21.9%) in T2, 6 (18.8%) in T3 and 12 (37.5%) in T4 at the time of diagnosis. In all cohort, 6 (18.8%) of them had lymph node metastasis. (**Table 1**) During the analysis, T1 and T2, T3 and T4 were placed together in two groups, and lymph node status were divided into two groups according to the presence of metastasis or not. T4A and T4B tumours were grouped together as T4. Twenty-one (65.6%) patients had free of surgical margins, and 9 (28.12%) positive margins, data of 2 (6.25%) cases missed. Grade was recorded in all patients. They were divided into two groups as being either low-grade (23 cases), including well and moderately differentiated tumors, or high-grade (9 cases), including poorly differentiated or undifferentiated tumors. For perineural invasion (PNI), 9 (28.1%) were positive, 13 (40.6%) were negative and 10 (31.3%) were missed. For lymphovascular invasion (LVI), 7 (21.9%) were positive, 20 (62.5%) were negative and 5 (15.6%) were missed. Four (12.5%) patients had facial paralysis at the time of first admission. All patients underwent a curative surgery. Among the whole cohort, 12 (37.5%) underwent total parotidectomy alone, 10 (31.25%) superficial parotidectomy, 10 (31.25%) total parotidectomy with neck dissection. Of these neck dissected 10 cases, 6 (18.75%) of theme underwent therapeutic and 4 (12.5%) elective dissection. Node involvement was present in 6 (18.75%) cases. Only patients who underwent definitive PORT were included in the study. For the cohort, RT indications were as follows: patients with T3-T4 tumor, and/or lymph node positivity, and/or perineural, and/or lymphovascular invasion, and/or positive surgical margins, and/or high-grade tumor, and/or extraglandular spread were considered high-risk patients and had one or more of these features were treated by RT. The median duration of RT was 44 days (range 39-52). For 18 (56.25%) patients, RT was applied only to the postoperative tumor bed, and for 14 (43.75%) patients, the neck region was also included in the RT treatment area. An average of 50 Gy (46-66 Gy) delivered to the neck region and 60 Gy (50-70 Gy) for the tumor bed.

Table 1. Demographic and pathologic features

Baseline characteristic	Total No. patients	Percent
Gender		
Female	18	56.2%
Male	14	43.7%
Grade		
Low-grade (1 or 2)	23	71.8%
High-grade (3 or 4)	9	28.1%
T stage		
T1or T2	14	43.7%
T3 or T4	18	56.2%
Lymph node status		
N0	26	81.2%
N+	6	18.8%
Margin		
Negative	21	65.6%
Positive	9	28.1%
Missed	2	6.2%
Perineural invasion		
Invasion –	13	40.6%
Invasion +	9	28.1%
Missed	10	31.3%
Lymphovascular invasion		
Invasion –	20	62.5%
Invasion +	7	21.9%
Missed	5	15.6%

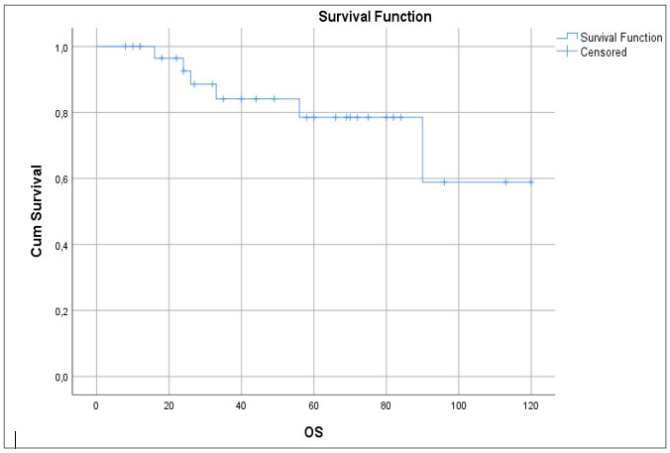
No patients received a chemotherapy for curative intent. First doctor visits were made 4-6 weeks after the end of treatments. Than, patients were followed-up every 3 months for 2 years, and every 6 months thereafter. Physical examination routinely performed for each single patients. Head-neck/thorax computed tomography scan was performed during the follow-up period, if necessary.

Survival Outcomes and Prognostic Factors

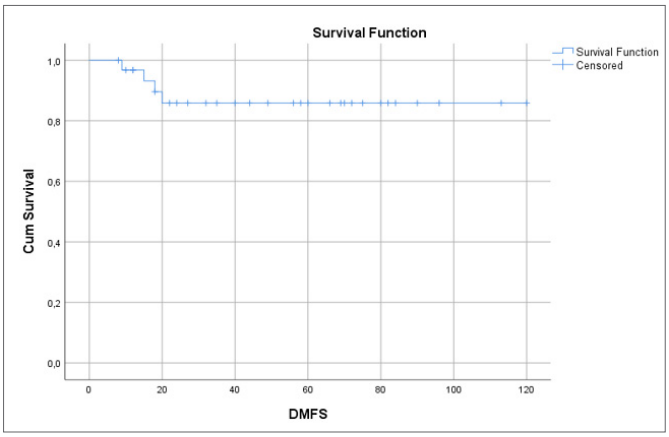
The 2-year and 5-year OS rates were 92.6% and 78.5% (**Graphic 1**), LRFS rates were 100% and 89.1%, DMFS rates were 85.9% and 85.9% (**Graphic 2**), respectively. Locoregional recurrence detected in 2 (6.25%), distant metastases detected in 4 (12.5%) patients. All distant metastases detected in the lungs. Locoregionally recurred patients both recurred locally, there was no regional recurrence in this cohort.

Univariate analysis showed that age, gender, margin status, T stage, facial nerve involvement, lymphovascular invasion, and perineural invasion were not significantly related to OS (all $p > 0.05$). Lymph node involvement ($p<0.013$) and grade ($p<0.006$) were the only significant prognostic factors for OS. In multivariate analysis, both lymph node involvement ($p<0.050$) and grade ($p<0.028$) remained the significant prognostic factors.

Univariate analysis revealed that gender ($p<0.018$) and lymph node involvement ($p<0.001$) were the only significant prognostic factors for LRFS. In multivariate analysis, neither were significant prognostic factors.



Graphic 1. Overall Survival curve



Graphic 2. Distant Metastases Free Survival curve

Univariate analysis revealed that grade ($p<0.001$) were the only significant prognostic factor for DMFS. Thus, multivariate analysis was not performed.

DISCUSSION

In most series, parotid gland ACCs were detected more in women than in men (6). Duzova et al. (7) reported a female dominancy for ACCs histology. Similarly, in the presented cohort, females (56.25%) are more affected than males (43.75%). This tumor affect a wide age range, from younger to older people. It can even be detected in pediatric age groups (8). Most ACCs appear between the ages of 40-49 years (6). In our cohort, age ranged from 35 to 80 years and most of theme were in fourth (34.37%) and fifth (34.37%) decades.

Efforts related to histological grading have not reached a definitive conclusion and are still a controversial issue. Features often associated with more aggressive tumors include frequent mitosis, focal necrosis, neural invasion, pleomorphism, infiltration, and stromal hyalinization. In ACCs, cases of from low- to high-grade de-differentiation have been reported. These cancers are defined with cytological pleomorphism, accrued proliferative and mitotic activity, proliferation indices and having a poor prognosis (9). Currently, It is reported

that up to 35% of ACCs, which were wrongly thought to be a benign tumor in the past, are high-grade. This topic is too important, predictably, since high-grade ACCs are related with advanced disease, a higher incidence of recurrence, distant metastasis, and worse prognosis (10). In the present study, high-grade tumor associated with poorer OS both for univariate and multivariate analysis and it seemed to be related with distant metastasis. In a retrospective study by Fang et al. (11) analyzed the medical records of 144 patients and they reported high-grade and intraparotid lymph node positivity as an indepent prognosticators in patients with ACCs histology. In this study, female gender (57.6%) was dominant and the mean age of the study population was 54.8 years as similar to our study. High mitotic rate, high-grade transformation, close and involved surgical margins, and necrosis were negative prognosticators, according to the results of a recently reported Dutch study of 89 ACCs cases treated and retrospectively reviewed between 1979 and 2016. In conclusion, they suggested that due to the relatively high incidence of high-grade transformation (21%) in ACCs and the low accuracy of cytology, elective neck dissection may be considered as part of standard treatment (12).

Various studies have previously examined the rate of lymph node metastasis in major salivary gland acinar cell carcinoma and have reported varying results. In a retrospective review of 66 patients from four different institutions examining nodal metastases in acinar cell carcinoma of the parotid gland, a metastasis rate of 34.3% was found in cases with neck dissection, and the overall incidence for the entire group of 66 diseases was 18.2% (13). A relatively small retrospective study from a single institution examined 14 cases and showed a 9% lymph node metastasis rate (14). Nodal metastases of ACCs reported in the literature ranges from 0% to 43%; The rate of occult regional metastases ranges from 0% to 13% (15). In the present study, lymph node involvement was present in 6 (18.75%) cases. This rate was relatively high in the current study, as the population of our study consisted of patients receiving RT, and patients who were candidates for RT were high-risk patients. Additionally, local recurrence developed in 2 patients. Remarkably, both of these two patients consisted of T4 and node positive patients. Lymph node positivity was found to be a prognostic factor affecting both survival and recurrence. Similiarly, in a large retrospective study of 255 patients with major salivary gland carcinomas in Brazil, clinical stage, positive lymph nodes, facial palsy, and invasion of adjacent structures were found to be predictors of distant metastases (16).

Margin status appears to play an important role in survival in AciCC patients. Clear margin is a positive prognostic factor for survival (12). In a retrospective case series with medical record review, patients treated surgically between the years from 2000 to 2014 for ACCs of the parotid gland were identified from an institutional database and 45 patients were included for analysis. They aimed to evaluate the effect of RT, particularly in patients with negative but close margins (≤1 mm) and no other high-risk histopathological factors. In conclusion, they stated that RT was unnecessary in patients with ACCs of the parotid gland with close (≤1 mm) but negative surgical margins and no other high risk factors (17).

Unlike many studies, in our cohort, perineural invasion and lymphovascular invasion are not rare features in ACCs, with an incidence of 28.1% and 21.9%, respectively. However, similar higher incidences were reported by Gomez et al. (23% vs 8.6%) (18). Despite this relatively low incidence of both features, their presence adversely affects survival, as in other salivary gland carcinomas (18,19). In the current study, neither perineural invasion nor lymphovascular invasion was a negative prognosticator (OS, LRFS and DMFS $p > 0.05$).

Surgery remains the mainstay of treatment in parotid gland ACCs. The necessity of adjuvant RT to whom in this patient group is still a controversial issue today. Although disease recurrences or distant metastases may be seen in some patients with parotid gland ACCs, most of the population with this disease has a more benign course and can be cured mostly with surgical treatment alone. Spafford et al. (20) reported recommendations regarding with adjuvant RT in ACCs in 1991. They recommended RT as an adjuvant treatment in addition to surgery in patients with the following features: recurrent disease, suspicious or surgical margins positivity, tumor adjacent to the facial nerve or facial nerve involvement, deep lobe involvement, lymph node metastases, extraglandular extension and tumors size greater than 4 cm. In a retrospective study published in 2018, Zenga et al. (17). aimed to illuminate a confusing issue. As we mentioned above, they questioned the role of RT in patients with ACCs with close (≤1 mm) surgical margins who had no other high risk factors and stated that RT was unnecessary in this group of patients. In another retrospective study published in 2018, Greig et al. (21) recommended adjuvant RT in high-grade tumors and in cases of surgical margin proximity or positivity. If we need to emphasize in line with the results of our study, there is a need for additional treatment options to adjuvant RT because the survival rate is lower in those with high grade and node positivity. Recent advances in drug studies have led to major changes in many treatment guidelines. In the era of immunotherapy, it is hoped that in the near future, drugs that will positively affect the prognosis in these tumor subgroups will be on the agenda.

The limitations of the present study should be acknowledged. First, there is an inherent bias in retrospective studies. Second, the sample size was relatively small, possibly reducing statistical power; Third, the entire patient population consisted of relatively high-risk patients undergoing adjuvant RT; therefore, studies with larger sample sizes are needed to confirm the results.

CONCLUSION

In ACCs of the parotid gland, high-grade histology and node positivity are independent variables that affect OS. Because survival is lower in these patient groups, it is imperative to explore other treatment options in addition to adjuvant RT. Multicenter, randomized studies are needed on this subject.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study protocol was reviewed and approved by the Clinical Research Ethics Committee (Date: 12/04/2022, Decision No: 2022/176).

Informed Consent: Written informed consents of all patients were acquired prior to treatment.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

Financial Disclosure: The authors declared that this study has received no financial support.

Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

REFERENCES

1. Khelfa Y, Mansour M, Abdel-Aziz Y, et al. Relapsed acidic cell carcinoma of the parotid gland with diffuse distant metastasis: case report with literature review. Med High Impact Case Rep 2016; 4: 2324709616674742.
2. Son E, Panwar A, Mosher C, et al. Cancers of the major salivary gland. J Oncol Pract 2018; 14: 99–108.
3. Godwin JT, Foote FW, Frazell EL. Acinic cell adenocarcinoma of the parotid gland; report of twenty-seven cases. Am J Pathol 1954; 30: 465–77.
4. Buxton RW, Maxwell JH, French AJ. Surgical treatment of epithelial tumors of the parotid gland. Surg Gynecol Obstet 1953; 97: 401–16.
5. Al Zaher N, Obeid A, Al-Salam S et al. Acinic cell carcinoma of the salivary glands: a literature review. Hematol Oncol Stem Cel Ther 2009; 2: 259–64.
6. Cavaliere M, De Luca P, Scarpa A et al. Acinic cell carcinoma of the parotid gland: from pathogenesis to management: a literature review. Eur Archi Otorhinolaryngol 2020; 277: 2673-9.
7. Düzova M, Turkan TS. Survival outcomes and prognostic factors in salivary gland cancers treated by surgery and adjuvant radiotherapy. Namık Kemal Tıp Derg 2021; 9: 108-15.
8. Bianchini C, Ciorba A, Stomeo F, et al. A case of parotid acinic cell carcinoma in a young boy. Brazilian J Otorhinolaryngol 2011; 77: 402.
9. Thompson L. World Health Organization classification of tumours: pathology and genetics of head and neck tumours. Ear Nose Throat J 2006; 85: 74.
10. Vander Poorten V, Triantafyllou A, Thompson LDR, et al. Salivary acinic cell carcinoma: reappraisal and update. Eur Archi Otorhinolaryngol 2016; 273: 3511-31.
11. Fang Q, Wu J, Du W, et al. Predictors of distant metastasis in parotid acinic cell carcinoma. BMC Cancer 2019; 19: 1-6.
12. van Weert S, Valstar M, Lissenberg-Witte B, et al. Prognostic factors in acinic cell carcinoma of the head and neck: The Amsterdam experience. Oral Oncology 2022; 125: 105698.
13. Grasl S, Janik S, Grasl M, et al. Nodal metastases in acinic cell carcinoma of the parotid gland. J Clin Med 2019; 8: 1315.
14. Rodriguez-Cuevas S, Labastida S, Baena L, et al. Risk of nodal metastases from malignant salivary gland tumors related to tumor size and grade of malignancy. Eur Archi Otorhinolaryngol 1995; 252: 139-42.
15. Napier SS, Herron BT, Herron BM. Acinic cell carcinoma in Northern Ireland: a 10-year review. British J Oral Maxillofac Surg 1995; 33: 145-8.
16. Mariano FV, Da Silva SD, Chulan TC, et al. Clinicopathological factors are predictors of distant metastasis from major salivary gland carcinomas. Int J Oral Maxillofac Surg 2011; 40: 504-9.
17. Zenga J, Parikh AS, Emerick KS, et al. Close margins and adjuvant radiotherapy in acinic cell carcinoma of the parotid gland. JAMA Otolaryngol Head Neck Surg 2018; 144: 1011-6.
18. Gomez DR, Katabi N, Zhung J, et al. Clinical and pathologic prognostic features in acinic cell carcinoma of the parotid gland. Cancer 2009; 115: 2128-37.
19. Vander Poorten V, Triantafyllou A, Thompson LDR, et al. Salivary acinic cell carcinoma: reappraisal and update. Eur Archi Otorhinolaryngol 2016; 273: 3511-31.
20. Spafford PD, Mintz DR, Hay J. Acinic cell carcinoma of the parotid gland: review and management. J Otolaryngol 1991; 20: 262-6.
21. Greig SR, Chaplin JM, McIvor NP, et al. Acinic cell carcinoma of the parotid gland: Auckland experience and literature review. ANZ J Surg 2008, 78: 754-8.

Original Article

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Clinicopathological evaluation of our patients with ultrasound assisted percutaneous kidney biopsy

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ABSTRACT

Aim: This study aims to determine the frequency of kidney diseases based on histological diagnosis and to evaluate the relationship between clinical and histopathological findings in patients undergoing percutaneous kidney biopsy for various indications.

Material and Method: In this cross-sectional study, demographic, anthropometric and laboratory data of the patients were obtained retrospectively from medical files and computer records. Biopsy indications and histopathological diagnoses (primary glomerular diseases, secondary glomerular diseases, tubulointerstitial diseases and other causes) of the patients were examined.

Results: Of 103 patients, 57 (55.3%) were male and 46 (44.7%) were female. The mean age of the patients was 44.67±15.29 years. The most common biopsy indication was hematuria+proteinuria+renal dysfunction (n=28, 27.2%). The most common pathology in histopathological diagnoses was primary glomerular diseases (56.3%), the most common diagnosis was IgAN (n=16, 15.5%). Tubulointerstitial diseases were seen more frequently in the 60 years and older group. (n=4, 25%). The most common cause of secondary glomerulonephritis was AA amyloidosis. The number of tubular disorders increased with advanced age.

Conclusion: In our center, renal biopsy was performed most frequently with the combination of proteinuria, hematuria, and renal dysfunction. The most common histopathological result was primary glomerulonephritis, in which IgAN took the first place.

Keywords: glomerulonephritis, histopathological diagnoses, renal biopsy

INTRODUCTION

Percutaneous kidney biopsy has a crucial role in diagnosing kidney diseases and guiding treatment in nephrology practice. It is the gold standard method in the diagnosis of renal parenchymal diseases, in predicting the course of the disease and in detecting the need for treatment (1). Today, it is performed with automatic or semi-automatic needles under the guidance of ultrasonography (USG), and the incidence of major complications is fairly low (2).

The most common indications for percutaneous kidney biopsy are nephrotic syndrome, nephritic syndrome, isolated microscopic hematuria, asymptomatic urinalysis abnormalities, acute kidney injury, suspected transplant kidney rejection, multisystemic diseases with renal involvement, and unidentified loss of kidney function (3). Diagnoses identified in kidney biopsies differ corresponding to the age, gender of the patients, whether they have native or transplanted kidneys, biopsy indications of centers, and race and geographical distribution (4).

This study purposes to determine the frequency of kidney diseases based on histological diagnosis and to assess the relationship between clinical and histopathological findings in patients who underwent percutaneous kidney biopsy for various indications in our center.

MATERIAL AND METHOD

This study was initiated after the approval of the Dışkapı Yıldırım Beyazıt Training and Resarch Hospital Ethics Committee (Date: 11/01/2021, Decision No: 102/01), and all procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki.

In this retrospective and cross-sectional study, the frequency of kidney diseases identified in patients who had percutaneous kidney biopsy with different indications and the relations between clinical and histopathological findings were examined. Informed consent form was obtained from all patients before the procedure.

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The Demographic, anthropometric, and lab data of the subjects were acquired retrospectively from medical files and computer registries.

Biopsy indications were microscopic hematuria (>2 erythrocytes per amplification field in urine sediment examination), proteinuria (>150 mg/day), renal dysfunction (unidentified loss of glomerular filtration rate), and post-treatment control (relapsed or treatment-resistant disease).

Percutaneous kidney biopsies were performed by the Interventional Radiology Unit or our nephrology team, using semi-automatic 16G and 18G biopsy needles, in the prone position, with at least 2 samples taken. The samples were placed on gauze impregnated with saline and delivered to Hacettepe University of Medicine Pathology Department within one hour in Petri dishes. A 2–4 mm long renal cortical tissue sample was reserved for examination and frozen sections were taken from these samples with a cryostat. IgG, IgA, IgM, C3, C1q, fibrinogen, kappa, and lambda dyes were applied by immunofluorescence method and evaluated with special filters in the immunofluorescence microscope. The remaining biopsy specimen was fixed with 10% formaldehyde. After routine tissue follow-up, 4µm-thick sections of tissue samples embedded in paraffin blocks were stained with Hematoxylin & Eosin, Periodic Acid Schiff, Masson Trichrome, Congo Red, and Methenamine Silver dyes and evaluated under a light microscope.

Biopsy results were classified as 1-Primary glomerular diseases [immunoglobulin A nephropathy (IgAN), focal segmental glomerulosclerosis (FSGS), membranous glomerulonephropathy (MGN), minimal change disease (MCD), membranoproliferative glomerulonephritis (MPGN), crescentic glomerulonephritis (anti-neutrophil cytoplasmic antibodies (ANCA)-associated vasculitis and anti-glomerular basement membrane antibody glomerulonephritis), post-infectious glomerulonephritis (PIGN)], 2-Secondary glomerular diseases [AA amyloidosis, systemic lupus erythematosus nephritis, diabetic nephropathy, monoclonal immunoglobulin deposition disease (MIDD), vascular renal damage (thrombotic microangiopathies)]; 3-Tubulointerstitial diseases (acute tubulointerstitial nephritis) (ATIN), 4-Hereditary renal diseases, 5-Chronic changes and 6-Others (oxalate nephropathy, nephrocalcinosis, undetermined diagnoses).

Statistical Analysis

Statistical analyzes were performed using the 25th version of the Statistical Package for the Social Sciences software (SPSS Inc., Chicago, IL, USA). The distribution of numerical variables was assessed using the Kolmogorov-Smirnov test. Normally distributed continuous data were compared with Student's t-test, and the results of the tests were given as mean values±standard deviation (SD). Abnormally distributed data were compared with the Mann-Whitney U test and results were presented as median and interquartile range. Comparisons of categorical variables were made using the Chi-square test and Fisher's exact test; results were presented as numbers and percentages.

RESULTS

All adult patients (n=112) who had a renal percutaneous biopsy in our nephrology clinic between November 2018 and March 2020 were assessed for the study. Patients whose biopsy specimens were technically inadequate for histopathological assessment (n=3) and patients who had biopsy from a transplanted kidney (n=6) were omitted from the study. 103 patients were recruited for the study eventually. Of the patients included in this study, 57 (55.3%) were male and 46 (44.7%) were female. The mean age of the patients was 44.67±15.29 years. Of these, 42 were under the age of 40, 45 were between the ages of 40-60, and 16 were aged 60 and over. In the patient groups separated according to biopsy indications, the mean age of those who underwent biopsy with the indication of isolated renal dysfunction was 61.75±17.37 years, while the mean age of the other groups ranged from 35.65±15.20 to 49.00±17.26 years (**Table 1**).

Table 1. Age and gender distribution of patients according to biopsy indications

		Gender		
Biopsy indication	**n=103**	**Male (n=57)**	**Female (n=46)**	**Age**
Hematuria, proteinuria and kidney dysfunction	28 (27.2%)	19 (67.9%)	9 (32.1%)	49.00±17.26
Proteinuria and kidney dysfunction	26 (25.2%)	14 (53.8%)	12 (46.2%)	44.58±12.77
Isolated proteinuria	25 (24.3%)	11 (44%)	14 (56%)	43.88±11.70
Nephrotic range proteinuria	13 (12.6%)	9 (69.2%)	4 (30.8%)	11.90
Non-nephrotic proteinuria	12 (11.7%)	2 (16.7%)	10 (83.3%)	44.08±11.99
Hematuria and proteinuria	17 (16.5%)	11 (64.7%)	6 (35.3%)	35.65±15.20
Isolated kidney dysfunction	4 (3.9%)	1 (25%)	3 (75%)	61.75±17.37
Re-biopsy after treatment	3 (2.9%)	1 (33.3%)	2 (66.7%)	40.00±18.68

Categorical variables are shown as frequency and percentages.

The most common biopsy indication in our study population was hematuria+proteinuria+renal dysfunction (n=28, 27.2%). This indication was also

the most common among men (n=19, 33.3%). Other biopsy indications were proteinuria+renal dysfunction (n=26, 25.2%), isolated proteinuria (n=25, 24.3%), hematuria+proteinuria (n=17, 16.5%), isolated renal dysfunction (n=4, 3.9%) and post-treatment control (n=3, 2.9%) (**Table 1**). None of the patients underwent biopsy, with indications of either isolated hematuria or hematuria+renal dysfunction.

In the analysis in which histopathological diagnoses were grouped, primary glomerular diseases had the highest rate (n=58, 56.3%). Among these, the most common diagnosis was IgAN (n=16, 15.5%). The most common primary glomerular disease identified in men was IgAN (n=12, 21.1%), while in women it was FSGS (n=7, 15.2%). When the distribution is made according to age groups, IgAN in patients younger than 40 years of age (n=9, 21.4%), FSGS in the 41-59 age group (n=11, 24.4%), and ATIN in patients 60 years and older (n=4, 25%) were the most common pathologies (**Table 2**).

The histopathological distribution of the patients according to biopsy indications is shown in **Table 3**. Hematuria+proteinuria (n=5, 38.5%) for MGN, hematuria+proteinuria+kidney dysfunction (n=7, 43.8%) for IgAN, nephrotic range proteinuria (n=6, 40%) for FSGS, nephrotic range proteinuria (n=3, 60 %) for MCD. The most common indication (n=6, 85.7%) was hematuria+proteinuria+renal dysfunction in patients with the diagnosis of crescentic glomerulonephritis.

DISCUSSION

This study intends to estimate out the frequency of renal diseases based on histological diagnosis and to evaluate the relationship between clinical and histopathological findings in patients who underwent percutaneous kidney biopsy for various indications in our center.

We considered the data from 103 patients who went through percutaneous biopsy of their native kidney over a course of almost one and a half years. The mean age of the patients at the point of diagnosis was 44.67±15.29 years, and 55.3% were male. These results were comparable to the data in the studies of Piskinpaşa et al. (5) and O'Shaughnessy et al. (6).

Although the indications for kidney biopsy differ according to race, geography and centers, the most common indication reported to date is proteinuria (7-10). In our study, unlike other studies, the most common biopsy indication was hematuria+proteinuria+renal dysfunction. When we reevaluate the biopsy indications in our study, it is noteworthy that only 7 patients did not have proteinuria. Our most common indication for biopsy may have appeared to be different, as our classification was different from other studies.

According to the 2020 Registry report of the Turkish Society of Nephrology, the most common cause of chronic renal failure in patients undergoing dialysis after diabetes and hypertension is glomerulonephritis (11). Glomerular diseases are the leading diseases diagnosed by percutaneous kidney biopsy.

Table 2. Distribution of histopathological diagnoses by gender and age

		Gender		Age groups		
Diagnosis	**Total (n=103)**	**Male (n=57)**	**Female (n=46)**	**≤40 year old (n=42)**	**40-59 (n=45)**	**≥60 year old (n=16)**
IgAN	16 (15.5%)	12 (21.1%)	4 (8.7%)	9 (21.4%)	5 (11.1%)	2 (12.5%)
FSGS	15 (14.6)	8 (14%)	7 (15.2%)	3 (7.1%)	11 (24.4%)	1 (6.3%)
MGN	13 (12.6)	9 (15.8%)	4 (8.7%)	4 (9.5%)	7 (15. 6%)	2 (12.5%)
ATIN	10 (9.7%)	4 (7.0%)	6 (13.0%)	5 (11.9%)	1 (2.2%)	4 (25%)
Chronic changes	11 (10.7%)	7 (12.3%)	4 (8.7%)	4 (9.5%)	6 (13.3%)	1 (6.3%)
Crescentic glomerulonephritis	7 (6.8%)	6 (10.5%)	1 (2.2%)	1 (2.1%)	3 (6.7%)	3 (18.8%)
AA amyloidosis	7 (6.8%)	2 (3.5%)	5 (10.9%)	3 (7.1%)	3 (6.7%)	1 (6.3%)
MCD	5 (4.9%)	3 (5.3%)	2 (4.3%)	2 (4.8%)	3 (6.7%)	-
Hereditary renal diseases	4 (3.9%)	1 (1.8%)	3 (6.5%)	2 (4.8%)	2 (4.4%)	-
Lupus nephritis	4 (3.9%)		4 (8.7%)	4 (9.5%)	-	-
Diabetic nephropaty	3 (2.9%)	2 (3.5%)	1 (2.2%)	1 (2.4%)	2 (4.4%)	-
MPGN	2 (1.9%)	1 (1.8%)	1 (2.2%)	2 (4.8%)	-	-
Nephrocalcinosis	1 (1.0%)	1 (1.8%)	-	1 (2.4%)	-	-
Oxalate nephropathy	1 (1.0%)	-	1 (2.2%)	-	-	1 (6.3%)
MIDD	1 (1.0%)		1 (2.2%)	-	1 (2.2%)	-
TMA	1 (1.0%)	-	1 (2.2%)	-	-	1 (6.3%)
PIGN	1 (1.0%)	-	1 (2.2%)	-	1 (2.2%)	-
Undetermined diagnoses	1 (1.0%)	1 (1.8%)	-	1 (2.4%)	-	-

Categorical variables are shown as frequency and percentages. Abbreviations: IgAN: immunoglobulin A nephropathy, FSGS: focal segmental glomerulosclerosis, MGN: membranous glomerulophropathy, ATIN: acute tubulointerstitial nephritis, AA: amyloid A, MCD: minimal change disease, MPGN: membranoproliferative glomerulonephritis, MIDD: monoclonal immunoglobulin deposition disease, TMA: thrombotic microangiopathy, PIGN: postenfectious glomerulonephritis

Table 3. Distribution of histopathological diagnoses of patients according to biopsy indications

	Biopsy Indication							
	Isolated proteinuria							
Diagnoses	**Non-nephrotic proteinuria**	**Nephrotic range proteinuria**	**Hematuria and proteinuria**	**Isolated kidney dysfunction**	**Proteinuria and kidney dysfunction**	**Hematuria, proteinuria and kidney dysfunction**	**Re-biopsy after treatment**	**Total**
MGN	2 (15.4%)	3 (23.1%)	5 (38.5%)	-	2 (15.4%)	1 (7.7%)	-	13 (100%)
IGAN	2 (12.5%)	-	3 (18.8%)	-	4 (25%)	7 (43.8%)	-	16 (100%)
FSGS	3 (20%)	6 (40%)	2 (13.3%)	-	3 (20%)	1 (6.7%)	-	15 (100%)
MCD	-	3 (60%)	-	-	2 (40%)	-	-	5 (100%)
MPGN	-	-	1 (50%)	-	-	1 (50%)	-	2 (100%)
Crescentic glomerulonephritis	-	-		-	-	6 (85.7%)	1 (14.3%)	7 (100%)
Hereditary renal diseases	1 (25%)	-	2 (50%)	-	-	1 (25%)	-	4 (100%)
ATIN	-	-	2 (20%)	2 (20%)	4 (40%)	2 (20%)	-	10 (100%)
AA amyloidosis	1 (14.3%)	-	-	-	3 (42.9%)	3 (42.9%)	-	7 (100%)
Lupus nephritis	1 (25%)	-	1 (25%)	-	-	1 (25%)	1 (25%)	4 (100%)
Diabetic nephropaty	1 (33.3%)	1 (33.3%)	-	-	1 (33.3%)	-	-	3 (100%)
MIDD	-	-	-	-	-	1 (100%)	-	1 (100%)
TMA	-	-	-	1 (100%)	-	-	-	1 (100%)
PIGN	-	-	-	-	-	1 (100%)	-	1 (100%)
Chronic changes					7 (63.6%)	3 (37.3%)	1 (9.1%)	11 (100%)
Undetermined diagnoses	-	-	1 (100%)		-	-	-	1 (100%)
Oxalate nephropathy	-	-	-	1 (50%)	-	-	-	1 (100%)
Nephrocalcinosis	1 (50%)	-	-					1 (100%)

Categorical variables are shown as frequency and percentages. Abbreviations: IgAN: immunoglobulin A nephropathy, FSGS: focally segmental glomerulosclerosis, MGN: membranous glomerulophropathy, ATIN: acute tubulointerstitial nephritis, MCD: minimal change disease, MPGN: membranoproliferative glomerulonephritis, MIDD: monoclonal immunoglobulin deposition disease, TMA: thrombotic microangiopathy, PIGN: postenfectious glomerulonephritis AA: amyloid A

We detected glomerular disease in approximately 70% of all cases in our study group. 56.3% of glomerulonephritis was due to primary causes and 13.6% to secondary causes. Among primary GN, the most common cause was IgAN (15.5%). This result was similar to the results previously reported in Czechia (9), Italy (4), France (12), England (13), USA (14), China (15), Korea (16) and our country (17). According to the very recently published KDIGO Clinical Practice Guidelines for Glomerular Diseases, IgAN is also the most common primary glomerular disease worldwide (18).

It was observed that there were differences in the pathological subtypes of glomerulonephritis according to age groups. The dominance of IgAN in the age group of 40 and below, of FSGS in the age group of 41-59, and of ATIN in the group of 60 and above was remarkable. In our analysis, as in similar studies (19), it was observed that the rate of renal tubular diseases increased while the rate of glomerular disease decreased as the mean age of the patients increased. The reason ATIN is frequently observed in the 60-year-old and older group may be the prevalence of over-the-counter drugs (especially analgesics) and herbal treatment used in this age group (20). In our study, data collection and analysis were not carried out in this direction. In the future, analyses that include data on drugs used and co-morbidities may provide clearer results.

In our study, the most common biopsy indication in patients diagnosed with MGN was hematuria+proteinuria. Although nephrotic syndrome is frequently seen in MGN, it should be kept in mind that 30-40% of it is accompanied by hematuria. (21).

In our study, AA amyloidosis was the first among secondary GNs. Although studies reporting lupus nephritis among the most common causes draw attention (12,22,23), there are also studies reporting AA amyloidosis (24). The geographic prevalence differences of diseases such as Familial Mediterranean Fever causing AA amyloidosis may contribute to these heterogeneous results.

The incidence of diabetic nephropathy may vary depending on the differences in kidney biopsy indications in diabetic patients. In our center, biopsy is not performed in diabetic cases with typical features of diabetic nephropathy, such as long-term DM, presence of DM-related microvascular complications such as diabetic retinopathy and neuropathy, and documented microalbuminuria-macroalbuminuria process. This approach may explain our low rate of diabetic nephropathy.

Study Limitations

The first is the evaluation of biopsy specimens only by light microscopy and immunofluorescence examination. Electron microscopic examination could not be performed. Secondly, the collection of cases from a single center resulted in a limited sample size.

CONCLUSION

In our center, the most common indication for kidney biopsy was proteinuria+hematuria+renal dysfunction, the most common primary glomerulopathy was IgAN, and the most common secondary glomerulopathy was AA amyloidosis. Tubular diseases were more common in advanced ages. We expect our findings will help us to build up faster and more accurate approaches to the target patient group.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was initiated with the approval of the Dışkapı Yıldırım Beyazıt Training and Research Hospital Ethics Committee (Date: 11.01.2021, Decision No: 102/01).

Informed Consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

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Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

REFERANCE

1. Korbet SM. Percutaneous renal biopsy. Semin Nephrol 2002; 22: 254-67.
2. Topham PS, Chen Y.: Renal Biopsy. In: Investigation of Renal Disease, Feehally J. Floege J. Tonelli M. Johnson RJ (eds). Comprehensive Clinical Nephrology, 6th ed. China: Elsevier; 2019: 72-9
3. Stratta P, Canavese C, Marengo M, et al. Risk management of renal biopsy: 1387 cases over 30 years in a single centre. Eur J Clin Invest 2007; 37: 954-63.
4. Gesualdo L, Di Palma AM, Morrone LF, Strippoli GF, Schena FP. Italian Immunopathology Group, Italian Society of Nephrology. The Italian experience of the national registry of renal biopsies. Kidney Int 2004; 66: 890-4.
5. Pişkinpaşa S, Dede F, Akoğlu H, et al. Böbrek biyopsilerinin klinikopatolojik değerlendirmesi: Tek merkez deneyimi. Turk Neph Dial Transpl 2012; 21: 167-72.
6. O'Shaughnessy MM, Hogan SL, Poulton CJ, et al. Temporal and demographic trends in glomerular disease epidemiology in the southeastern United States, 1986-2015. Clin J Am Soc Nephrol 2017; 12: 614-23.
7. Rivera F, López-Gómez JM, Pérez-García R. Spsnish Registry of Glomerulonephritis. Frequency of renal pathology in Spain 1994-1999. Nephrol Dial Transplant 2002; 17: 1594-602.
8. Rychlík I, Jancová E, Tesar V, et al. The Czech registry of renal biopsies. Occurrence of renal diseases in the years 1994-2000. Nephrol Dial Transplant 2004; 19: 3040-9.
9. Schena FP. Survey of the Italian Registry of Renal Biopsies. Frequency of the renal diseases for 7 consecutive years. The Italian Group of Renal Immunopathology. Nephrol Dial Transplant 1997; 12: 418-26.
10. Heaf J, Løkkegaard H, Larsen S. The epidemiology and prognosis of glomerulonephritis in Denmark 1985-1997. Nephrol Dial Transplant 1999; 14: 1889-97.
11. National Nephrology, Dialysis and Transplantation Registry Report of Turkey 2020. Türk Nefroloji Derneği Yayınları Ankara 2021.
12. Simon P, Ramee MP, Boulahrouz R, et al. Epidemiologic data of primary glomerular diseases in western France. Kidney Int 2004; 66: 905-8.
13. Hanko JB, Mullan RN, O'Rourke DM, McNamee PT, Maxwell AP, Courtney AE. The changing pattern of adult primary glomerular disease. Nephrol Dial Transplant 2009; 24: 3050-4.
14. Swaminathan S, Leung N, Lager DJ, et al. Changing incidence of glomerular disease in Olmsted County, Minnesota: a 30-year renal biopsy study. Clin J Am Soc Nephrol 2006; 1: 483-7.
15. Hou JH, Zhu HX, Zhou ML, et al. Changes in the spectrum of kidney diseases: an analysis of 40,759 biopsy-proven cases from 2003 to 2014 in China. Kidney Dis 2018; 4: 10-9.
16. Chang JH, Kim DK, Kim HW, et al. Changing prevalence of glomerular diseases in Korean adults: a review of 20 years of experience. Nephrol Dial Transplant 2009; 24: 2406-10.
17. Ecder SA, Kılıçaslan I, Ecder T ve ark. Beşyüzonüç böbrek biyopsisinin klinikopatolojik açıdan değerlendirilmesi. J Ist Faculty Med 2005; 68: 43-5.
18. Kidney Disease: Improving Global Outcomes (KDIGO) Glomerular Diseases Work Group. KDIGO 2021 clinical practice guideline for the management of glomerular diseases. Kidney Int 2021; 100: 1-276
19. Aydın, E, Yılmaz Aydın, F, Yılmaz, E, Alabalık, U. Böbrek biyopsilerinin histopatolojik değerlendirilmesi: tek merkez yedi yıllık deneyim. Dicle Tıp Derg 2020; 47: 417-22
20. Yılmaz T, Alp A, Akdam H. Böbrek biyopsisi yapılan olgularımızın retrospektif genel değerlendirilmesi ve histopatolojik alt gruplarının incelenmesi. Turk Neph Dial Transpl 2014; 23: 185-95.
21. Salant JD, Cattran CD: Membranous Nephropathy. In: Glomeruler Disease, Feehally J. Floege J (eds.). Comprehensive Clinical Nephrology, 6th ed. China: Elsevier; 2019: 240-53
22. Zhou FD, Shen HY, Chen M, et al. The renal histopathological spectrum of patients with nephrotic syndrome: an analysis of 1523 patients in a single Chinese centre. Nephrol Dial Transplant 2011; 26: 3993-7.
23. Akarsu Ö, Aytuğ F, Yavuz A, et al. Hastanemiz nefroloji kliniğinde böbrek biyopsisi yapılan olguların özelliklerinin değerlendirilmesi. Turk Neph Dial Transpl 2016; 25: 245-50.
24. Akın D, Sehmus Ö, Danış D: 2001-2007 döneminde nefrotik sendromlu 152 hastanın analizi. Turk Neph Dial Transpl 2008; 17: 74-6.

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Pharyngcutaneous fistula after total laryngectomy: treatment modalities and our experiences

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ABSTRACT

Aim: The aim of this study is to share our experience and treatment modalities in pharyngocutaneous fistulas that can be seen after primary and post-radiotherapy salvage total laryngectomy.

Material and Method: The records of patients who underwent total laryngectomy in our clinic between February 2012 and December 2021 were reviewed retrospectively. The patients' age, preoperative radiotherapy history, postoperative fistula development, gastrostomy opening, treatment modality, and hospital stay were examined.

Results: A total of 115 patients, 81 of whom were primary and 34 of whom had a history of radiotherapy, were evaluated in the study. Pharyngocutaneous fistula developed in a total of 20 patients in 10 primary patients and 10 patients with a history of preoperative radiotherapy. The mean hospital stay was 10 days for patients who did not develop fistula. The mean hospital stay was 27 days between days 21 and 36 in primary patients with fistula, and it was 46 days between days 34 and 68 in patients with a history of radiotherapy who developed fistula. While only 1 of the primary patients underwent reconstruction with a pectoralis major muscle flap because the fistula did not close despite local dressing and local skin flaps, this number was 7 in patients with a history of radiotherapy.

Conclusion: The presence of a history of radiotherapy before total laryngectomy increases the risk of laryngocutaneous fistula development, increases the need for myocutaneous flaps, and increases the length of hospital stay, resulting in increased comorbidity. Pharyngocutaneous fistula development is neither an important comorbidity nor religion.

Keywords: Total laryngectomy, radiotherapy, salvage laryngectomy, pharyngocutaneous fistula

INTRODUCTION

About 0.8% of all new cancer cases in the world and 0.6% of all cancer deaths occur in patients with laryngeal cancer (1). The incidence of Laryngeal Cancer (LC) has been decreasing 2.4% each year for the last 10 years with the decrease in tobacco use (1). LC occurs more frequently with among men and advancing age. LC is 5 times more common in men than women. The median age of diagnosis for patients with LC is 65 years and the median age at death is 68 years. The major risk factor for laryngeal cancer is smoking; other risk factors include human papillomavirus infection, laryngopharyngeal reflux, alcohol and environmental or occupational exposures. The incidence of LC among women has increased with the increase in smoking in women over past decades. The role of human papillomavirus as a risk factor for laryngeal cancer in young nonsmokers is currently under investigation (2).

In addition to surgical treatment in laryngeal tumors, radiotherapy (RT) in early-stage tumors is an effective treatment method in the treatment of laryngeal cancers. When the literature is examined, it has been shown that the oncological results of RT and cordectomy are similar in the treatment of early-stage tumors (3). From a functional point of view, the presence of vocal ligaments, absence of tissue loss and better sound quality make radiotherapy advantageous (3).

Primary tumor recurrence or persistence of disease after radiation therapy must be detected as early as possible. The role of surgical salvage in the event of disease recurrence should be considered even before initial treatment is selected (4,5). The high risks of wound healing complications should be considered in the planning for any surgery of the larynx after radiotherapy relative to surgery in an unirradiated neck

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(5). Therefore, patients who have poorly functioning larynx, suggestive of extensive T3 or T4a disease, and those with a tumor penetrating through the thyroid cartilage into surrounding soft tissues are not suitable for larynx preservation treatment approaches and should be recommended to undergo total laryngectomy (4).

In a study the incidence of pharyngocutaneous fistula (PCF) was 19% among primary total laryngectomy (TL), while it increased to 28.6% and 30.3% for patients receiving salvage TL after radiotherapy and chemoradiotherapy (6). In advanced stage tumors, partial surgery (in selected cases), TL (for T4a tumors and selected T3 patients) is a standard treatment method (7).

MATERIAL AND METHOD

This study was approved by the University of Health Sciences Ankara Oncology Training and Research Hospital Clinical Studies Ethics Board (Date: 2022-03, Decision No: 70). All procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki.

The records of a total of 115 patients who underwent TL f(70.4%) primary and 34 (29.6%) salvage due to recurrence after radiotherapy, in the ENT clinic of Ankara Oncology Training and Research Hospital were retrospectively analyzed. Patients' age, preoperative radiotherapy history, postoperative fistula development, approach to fistula, modality of treatment applied, and length of hospital stay were examined. The number of female patients was very low, so female patients were excluded from the study in order to avoid gender differences. Patients with Diabetes Mellitus and Thalassemia minor were excluded from the study because they may delay wound healing.

The surgical technique was standardised among all surgeons. TL included standard removal of the hyoid bone and infrahyoid muscles, pharyngeal closure was always performed in a single layer with a "I" shaped suture line, using 4-0 vicryl sutures. All patients were fed through a nasogastric tube in the postoperative period. Oral feeding was started on the 7th postoperative day in patients who did not develop FCF. Post-operative CRP follow-up was performed in all patients. Patients with evidence of postoperative infection were examined by the infectious diseases department and treated with appropriate antibiotics. During the physical examination; In cases where there was hyperemia on the neck skin, the flap did not fit, serohemorrhagic fluid comes from the neck suture lines, saliva and/or food content was observed in the hemovac drains, or if all these examination findings were normal, in cases where the patient's CRP and WBC values were increased, it was thought that it may be PCF, and some of the neck skin sutures were removed and an open neck dressing was applied. If hematoma, formula content, infected seroma, and saliva were observed in this dressing, the patient was considered to have PCF and was dressed. Consultations were requested from infectious diseases. If oral feeding was started, it was stopped and the dressing was continued twice a day. During this dressing; The space under the flap was cleaned, dead tissues were debrided, and the surrounding tissues were scraped and bled to increase granulation. The vascularity of the tissue was improved by scraping subcutaneously on the anterior line of the fistula. With this procedure, it was aimed to bond the living skin tissue with the esophagoplasty. Rifamycin ampoule and Triticum Vulgare aqueous extract and cream-impregnated sponge containing ethyleneglycol were placed on and around the flap skin, fistula area, and replaced with a new one at each dressing. For reconstruction, only one or more of the techniques of dressing, primary sturation, repair with local flap and repair with myocunous flap were applied. Endoscopic gastrostomy (PEG) was performed by the relevant department for patients whose fistula closure was not expected recently.

The age of the patients, the development of FCF, the length of hospital stay, the duration of feeding with nasogastric tube, the gastrostomy opening, the treatment approach applied to the fistula were evaluated. The data analysis was performed using the Statistical Package for Social Sciences (SPSS) for Windows, version 15. Chi-square and Mann-Whitney U tests were applied as statistical methods. A p-value of less than 0.05 was considered significant.

RESULTS

FCF developed in a total of 20 patients, including in primary 10 (12.3%) patients and in 10 (29.4%) patients who underwent salvage TL due to a history of preoperative RT. The primary patients were between 39 and 79 years old, with a mean age of 61.3±3.9 years. The age of the patients who relapsed after RT ranged from 38 to 76 years, with a mean age of 60.6±4 years. Nasogastric tube feeding was started in all patients on the 1st postoperative day, and all patients were fed with a nasogastric tube until oral feeding or gastrostomy feeding started. The mean hospital stay was 10 days for patients who did not develop fistula. The mean hospital stay was between 21 and 36 days in primary patients with fistula, and 27 days on average, while it was 34-68 days, with a mean of 46 days, in patients with a history of radiotherapy who developed fistula. In 7 of the primary patients, closure of the fistula was achieved in 6-20 days, with an average of 12 days, with dressing and debridement, whereas in only 1 of the patients with a history of RT, it closed in 19 days.

The fistula mouth was opened to the skin due to partial necrosis of the neck skin of 3 patients who were applied

primary TL and could not be successful with local dressing. Localized random skin flaps prepared from the neck skin were applied to these patients (A single lobe skin flap made from the lateral neck was closed over the fistula as the epidermal surface as the anterior esophageal wall, a similar flap was also prepared from the neck for the patients in need.). Fistula closed in 2 of 3 primary patients who underwent local skin flap.

Local flap was not considered in 3 of 9 patients with ongoing fistula problem and a history of RT due to fistula location and size. Local flap was applied to 6 patients and fistula closure was achieved in only 1 of these patients.

Pectoralis major (PM) myocutaneous flap was applied to primary 1 patient and 8 patients with a history of RT. While preparing the PM flap, the size of the skin island was determined by measuring according to the defect size. A circumferential incision was made around the skin graft. The pectoralis major muscle was cut from the inferior medial and lateral sides, similar to the skin size. The PM muscle was separated from the pectoralis minor and sternocostal muscles by blunt dissection. During dissection, the thoracoacromial artery was recognized and preserved. A tunnel was formed between the skin with the clavicle, and the skinned muscle, which was supported, was passed through the tunnel and reached the defective site. The flap was stitched over the defective area. Despite PM reconstruction, fistula persisted in only 2 patients with a history of RT, and contralateral pectoralis major myocutaneous flap was applied to these patients. Thus, fistula healing was achieved in all patients. Gastrostomy was performed by the relevant department in 8 primary patients with pharyngocutaneous fistula and all 10 patients with a history of rt. While the presence of gastrostomy was 15-28 days and an average of 21 days in primary patients, this period was 34-68 days and an average of 44 days in patients with a history of RT. Our findings are summarized in **Table 1.**

Table 1. Our data

	Primary Patients	Post-RT Salvage Patients
Median age	61.3±3.9	60.6±4
Number of patients	81	34
Patients Developing FCF	10	10
Length of hospital stay in the presence of PCF	27	46
Patients who had gastrostomy	8	10
FCFs closed with wound care	7	1
FCFs closed with local flap application	2	1
Patients with Pectoralis Major flap	1	8

When our patients were evaluated, no significant age difference was observed between primary patients and patients with a history of RT (p=0.07). Pharyngocutaneous fistula development was significantly higher in the group with a history of RT (p=0.03). Patients with a history of RT stayed significantly longer in hospital (P<0.001). Eight of 81 primary patients and 10 of 34 patients with a history of RT had gastrostomy, and the rate of gastrostomy was significantly higher in patients with a history of RT (p=0.011). In patients who underwent gastrostomy, the duration of gastrostomy was significantly longer in the group with a history of RT (p<0.001). Salvage patients with a history of RT required significantly more pectoralis major myocutaneous flaps (p=0.044).

DISCUSSION

With the evolution of nonsurgical organ preservation protocols for treatment of laryngeal and hypopharyngeal squamous cell carcinomas like RT, TL is increasingly performed as salvage procedure. Wound complications and FCF are important complications. Wound healing complications can have a multifactorial origin including previous chemoradiotherapy, low albumin , anemia, neck dissection, tumour stage and site (8).

The development of a FCF represents an important complication of TL that is usually self-limiting. Its management is thus mainly based on careful conservative treatment; however, at times, this complication further surgery is required (6).

The role of radiotherapy in the genesis of FCF has been extensively described, and some authors report that there is no significant associations (9). In a recent meta-analysis, Paydarfar et al. (10) repoted that, although preoperative radiotherapy is a significant relative risk of PCF formation, there was also heterogeneity of effects among studies; in fact, other RT associated variables such as radiotherapy time and dose frame between the end of radiation and surgery, did not demonstrate an increased.

Choosing a surgical versus a nonsurgical approach as the initial treatment for LC depends on individual patient factors such as comorbidities and age, the subsite of the tumor, the volume and extent of the primary tumor, and the presence of lymph node metastases or the probability of metastases. The choice of treatment is also influenced by involvement of the tumor location such us anterior commissure and the ability to achieve adequate endoscopic visualization. The presence of surgical and radiation oncologic expertise, along with adequate rehabilitative services, are fundamental considerations (4).

Other important factors deciding on treatment include vocal cord mobility, fixation, or impairment pretreatment voice and swallowing function, patient desires and lifestyle needs as related to the morbidity of treatment, and patient compliance (11).

TL is the standard treatment for advanced stage laryngeal tumor for patients not amenable to organ preservation regimens posed by extralaryngeal spread, extensive thyroid cartilage invasion or multiple and severe comorbidities. TL removes the entire larynx, paratracheal lymphatics, strap muscles, and the ipsilateral thyroid lobe or total thyroid excision in some cases (12). TL usually offers the best chances for cure, the consequences are the loss of native voice and a permanent tracheostoma. The most common complications of TL are wound infection and FCF, occurring in up to 50% of previously radiated patients. Therefore, vascularized tissue flaps to cover the reconstructed pharyngeal closure during salvage TL may be advised to decrease the risk and severity of fistulas preservation, salvage surgery offers yet a chance for cure (4,12). Careful consideration of the potential of successful salvage surgery to achieve loco-regional control is necessary before any laryngeal preservation approach with radiation or chemoradiation can be considered a viable alternative to primary surgery. Complete resection may be accomplished by TL, open partial laryngectomy or transoral laser microsurgery. Despite all the advances, the functional results of rehabilitation after partial laryngectomy, such as supraglottic laryngectomy, continue to be poor in patients who have received radiation before. Likewise, the results of supracricoid laryngectomy in surgical salvage on ultimate decannulation and local control should be interpreted with caution as the role for secondary partial laryngectomy becomes more clearly determined (12).

Because of the critical role of the larynx in voice and swallowing, the goals in the treatment of patients with T1 and T2 stage laryngeal cancers are cure from disease and preservation of laryngeal function to maximize quality of life. Although an approach combining multiple modalities such as surgery and radiation therapy or chemoradiation may be warranted on oncologic grounds, it can be togother high cost of added morbidity, including compromised functional outcomes of the larynx. Therefore, carefully selected patients with favorable early stage tumors should be considered for a single modality of therapy whenever possible without decreasing the chance for cure. Such single modality options include partial laryngectomy, transoral laser microsurgery, or radiation therapy alone may be considered standard treatment for laryngeal preservation for patients (4).

Busoni et al. (6) showed that the rate of PCF was 19% in primary total laryngectomy patients and %30.3 in post-RT laryngectomies, and showed that the history of RT increased the rate of PCF development. They also found significantly longer hospital stays. Similarly, in our study, the rate of PCF development, length of hospital stay, gastrostomy application rate and hospital stay, and major flap (pectoralis major myocutaneous flap) application rate were found to be high in patients with a history of RT.

CONCLUSION

While arranging the modality of treatment in patients diagnosed with laryngeal cancer, the increased comorbidity risks that may develop as a result of possible recurrences that may develop after RT should be kept in mind in patients who have had a chance of radiotherapy treatment. We think that the patient should be informed in detail about the possible results while the treatment plan is being made, and that a joint decision should be made with the physician, taking into account the possible risks and comorbidities.

ETHICAL DECLARATIONS

Ethics Committee Approval: This study was approved by the University of Health Sciences Ankara Oncology Training and Research Hospital Clinical Studies Ethics Board (Date: 2022-03, Decision No: 70).

Informed Consent: Since the study was designed retrospectively, informed consent was not obtained from the patients.

Conflict of Interest Status: The authors declared that there was no conflict of interest in this study.

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Author Contributions: All authors; declared that they participated in the design, execution, and analysis of the article and approved the final version.

REFERENCES

1. National Cancer Institute. SEER: cancer stat facts: larynx cancer. 2018. Available at: http://seer.cancer.gov/statfacts/html/laryn.html. Accessed February 10, 2018.
2. Megwalu UC, Sikora AG. Survival outcomes in advanced laryngeal cancer. JAMA Otolaryngol Head Neck Surg 2014; 140: 855–60
3. Mo HL, Li J, Yang X, et al. Transoral laser microsurgery versus radiotherapy for T1 glottic carcinoma: a systematic review and metaanalysis. Lasers Med Sci 2017; 32: 461-7
4. Forastiere AA, Ismaila N, Lewin JS, et al. Use of larynx preservation strategies in the treatment of laryngeal cancer: American Society of Clinical Oncology clinical practice guideline update. J Clin Oncol 2018; 36: 1143–69
5. Smee RI, Williams JR, Broadley K, Bridger GP. Early glottic carcinoma treated by radiotherapy: defining a populationfor surgical salvage. Laryngoscope 2013; 123: 171
6. M. Busonı, A. Deganello, O. Gallo. Pharyngocutaneous fistula following total laryngectomy: analysis of risk factors, prognosis and treatment modalities. Acta Otorhinolaryngol Ital 2015; 35: 400-5

7. Kirchner JA. Pathways and pitfalls in partial laryngectomy. Ann Otol Rhinol Laryngol 1984; 93: 301-5
8. Erdag MA, Arsianoglu S, Onal K, Songu M, Tuylu OA. Pharyngocutaneous fistula following total laryngectomy: multivariate analysis of risk factors. Eur Arch Otorhinolaryngol 2013; 270: 173-9
9. Emerick KS, Tomycz L, Bradford CR et al. Primary versus secondary tracheoesophageal puncture in salvage total laryngectomy following chemoradiation. Otolaryngol Head Neck Surg 2009; 140: 386–90
10. Deganello A, Gallo O, De Cesare JM, et al. Supracricoid partial laryngectomy as salvage surgery for radiation therapy failure. Head Neck 2008; 30 :1064-71
11. Gourin CG, Conger BT, Sheils WC, Biladeu PA, ColemanTA, Porubsky ES. The effect of treatment on survival in patients with advanced laryngeal carcinoma. Laryngoscope 2009; 119: 1312
12. Cohen JI, Clayman GL. Unit IV: laryngopharyngeal operations. In: Atlas of head & neck surgery. Philadelphia: Elsevier; 2011.

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Management of difficult bile duct stones with temporary plastic stent and ursodeoxycholic acid treatment: 5 years of experience

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ABSTRACT

Aim: The aim of the study was to investigate the effect of temporary plastic stenting and ursodeoxycholic acid (UDCA) treatment on difficult choledochal stones that cannot be removed by basic ERCP techniques in patients who applied to our clinic with occlusion and underwent endoscopic retrograde cholangiopancreatography (ERCP).

Material and Method: Patients were scanned retrospectively using the hospital database. Patients who underwent ERCP due to malignancy, biliary tract injury and benign strictures were excluded from the study. 61 patients who were not successful with basic ERCP techniques such as endoscopic sphincterotomy (EST) and mechanical lithotripsy (ML) were included in the study. 750 mg/day UDCA was given to the patients for three months and plastic stent was applied. After the treatment, ERCP was tried again.

Results: Among the patients who underwent stent+UDCA, three (4.9%) patients had perioperative bleeding, one (1.6%) patient had peroperative perforation, four (6.6%) patients had postoperative pancreatitis, and one (1.6%) patient had mortality. The mean hospital stay was 1.96±2.1 days. Post-procedure total bilirubin and direct bilirubin values were observed to be lower than before the procedure (respectively, $p<0.001$ and $p<0.001$). The reduction in common bile duct diameter and stone size was found to be statistically significant in patients who underwent two procedures (respectively, $p<0.001$ and $p<0.001$). Although the reduction in stone size was statistically significant in patients who underwent three procedures, the decrease in the diameter of the common bile duct was not significant (p=0,090).

Conclusion: In our study, temporary plastic stenting and UDCA treatment were shown to be beneficial in common choledochal stones that could not be removed with basic ERCP techniques in the first ERCP session.

Keywords: Endoscopic retrograde cholangiopancreatography, choledocholithiasis, endoscopic sphincterotomy, ursodeoxycholic acid and bile duct stent

INTRODUCTION

Nearly 7-12% of patients undergoing cholecystectomy for gallstones are found to have stones in the common bile duct. Endoscopic retrograde cholangiopancreatography (ERCP) is widely used in the treatment of common bile duct stones ranging in size from a few mm to 3 cm (1,2). Although 85-90% of common bile duct stones can be successfully removed with standard techniques such as endoscopic sphincterotomy (EST), endoscopic papillary balloon dilation (EPBD), and mechanical lithotripsy (ML), additional ERCP procedures or surgical intervention are required in some cases (3). Large (>1.5 cm), multiple, and cylindrical stones as well as stones that cannot be removed due to surgically altered anatomy, narrowed or angled distal bile ducts, and periampullary diverticulum are defined as bile duct stones difficult for ERCP (4). These patients need ERCP methods that require more advanced techniques and equipment such as extracorporeal shock wave lithotripsy (ESWL), electrohydraulic lithotripsy (EHL), laser lithotripsy (LL), and endoscopic papillary large balloon dilatation (EPLBD) (5,6). Even if these methods are known to be effective, their use is restricted in patients with comorbidities because they are not available in all centers, are technically more difficult, have higher costs, and have longer procedure times (7).

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In cases where advanced ERCP techniques could not be applied, temporary stenting was performed and difficult bile duct stones were removed with repetitive ERCPs. It is not entirely clear by what mechanisms temporary plastic stents play a role in the removal of stones, but it is assumed that they are caused by mechanical friction. It has been argued that the addition of ursodeoxycholic acid (UDCA) to temporary stenting is beneficial (8,9). Ursodeoxycholic acid treatment has been used for many years in the liver, biliary tract, and digestive system diseases. It has been shown to be beneficial especially in dissolving cholesterol gallstones and preventing cholestasis, which is seen as a risk factor in the formation of bile duct stones (10). In this study, it was investigated whether UDCA and temporary plastic stenting are beneficial in the treatment of difficult choledochal stones that cannot be removed with primary ERCP techniques.

MATERIAL AND METHOD

The study was initiated with the approval of the Bandırma Onyedi Eylül University Faculty of Medicine Clinical Researches Ethics Committee (Date: 09.05.2022, Decision No: 2022-67). In the study, 585 patients who underwent ERCP for obstructive jaundice in the General Surgery Department of Bandirma Onyedi Eylül University Faculty of Medicine between 2016-2021 were retrospectively analyzed.

Data Collection and Patient Selection

Patients who presented to our clinic with obstructive jaundice between the specified dates and underwent ERCP were scanned retrospectively using the hospital database. 55 patients who underwent ERCP due to malignancy, biliary tract injury, and benign strictures were excluded from the study. Stone extraction with EST and ML, technically applied in our clinic, was planned for 530 patients with common bile duct stones. Bile flow was achieved by successful stone extraction in 450 patients in the first ERCP session. A temporary plastic stent was placed in 61 patients and 750 mg/day UDCA treatment was initiated. In the remaining 19 patients, common bile duct cannulation could not be performed during ERCP. Twelve of these patients underwent surgical intervention, and 7 patients were referred to centers where advanced ERCP techniques could be applied, considering their general condition was not suitable for surgical intervention.

Pre-Operative Evaluation and ERCP Procedure

Before the procedure, all patients were evaluated in terms of anesthesia, and necessary medical treatments were planned to reduce mortality and morbidity. Oral intake was stopped 6-8 hours before the procedure and the patients were taken to the ERCP unit after prophylactic antibiotic administration. They were then placed in the left lateral decubitus and semi-prone position; topical local anesthetic spray [3-5 puffs (10 mg/puff) 10% lidocaine spray]was applied before the procedure. The procedures were performed under deep sedation [intravenous midazolam (0.05 mg/kg) and/or propofol (0.5 mg/kg) and/or fentanyl (0.5-1 µr/kg)]. After evaluating the esophagus and stomach, the second part of the duodenum was reached by passing through the pylorus. Following visualizing the ampulla vatery, the papillae facing position was provided. Selective bile duct cannulation was attempted by entering the sphincterotomy from the papilla orifice at the 11 o'clock orientation. In our clinic, guidewire supported cannulation is generally applied; however, in cases where cannulation cannot be performed in this way, selective cannulation is preferred with contrast-assisted cannulation and deep cannulation techniques. Similarly, the selective bile duct cannulation technique with guidewire support to the pancreas is also applied primarily in cases where bile duct cannulation cannot be performed and pancreatic cannulation can be performed. After the biliary tract is cannulated with the selective bile duct cannulation technique, the bile ducts are filled with contrast material and the biliary tract is visualized with C-arm scopy. After evaluating the bile duct pathologies, biliary sphincterotomy (EST) is performed by cutting the papilla between 11 o'clock and 1 o'clock directions with the help of a sphincterotomy and electrocautery. The upper border of the incision forms the point where the intraductal part of the papilla intersects with the duodenal wall. In cases where the bile duct cannot be cannulated selectively, the pre-cut method can be used. Biliary and pancreatic sphincters are exposed by cutting the papilla mucosa and submucosa starting from the orifice with a needle-pointed sphincterotomy or using the fistulotomy technique from the upper part of the orifice, and EST is performed after the bile duct is cannulated. Following that, the detected pathology is treated. Before or after the EST procedure, endoscopic papillary balloon dilatation (EPBD) and ML techniques can be applied for common bile duct stones. The ML consists of a mechanical lithotripter, a reinforced wire basket used to attach the stone to the bile duct, a metal sheath, and a handle in which the trapped stone and the basket are pulled back against the metal sheath, thus exerting a crushing force. Stone removal procedures with EPBD and ML techniques were planned for our patients. In case of failure, surgery was planned or a temporary plastic stent (8.5 Fr or 10 Fr) was placed to provide bile flow, and treatment was started with UDCA 750 m/day. Then, as long as there was no clinical deterioration, the ERCP procedure was repeated every three months. During this period, UDCA treatment was continued. In patients with clinical deterioration, surgical treatment or referral to centers where advanced ERCP techniques are performed was considered.

Follow-up

When the patients were evaluated in the recovery room after the procedure, some of them were discharged on the same day while some were taken to the service for follow-up purposes, according to their clinical status. Patients who were found to have abdominal pain during the physical examination were followed closely in terms of possible pancreatitis or perforation, and complete blood count, liver function tests, and amylase values were checked in the follow-ups.

Statistical Analysis

SPSS (Statistical Package for the Social Sciences) 25.0 package program was used for the statistical analysis of the data. Categorical measurements were expressed as numbers and percentages while continuous measurements as mean and standard deviation. Chi-square and Fisher's exact tests were used to compare categorical expressions. Shapiro-Wilk test was used to determine whether the parameters in the study showed a normal distribution. Mann Whitney U test was used for the parameters that did not show normal distribution. Wilcoxon ranks test was used to examine the differences between pre-and post-process values. The statistical significance level was considered as 0.05 in all tests.

RESULTS

In the study, a total of 585 ERCP patients were evaluated. 55 patients were directly excluded from the study because they were treated for malignancy, benign stenosis, and postoperative bile leakage. In the remaining 530 patients, stones were detected in the common bile duct, and stones were successfully removed in 450 (84.9%) in the first ERCP session. Twelve (2.26%) of 80 (15.1%) patients who were considered to have difficult common bile duct stones were scheduled for surgery, and seven patients (1.32%) were referred for advanced ERCP techniques. A temporary plastic stent was placed with ERCP in 61 patients (11.5%) and UDCA treatment was started. It was determined that the presence of concurrent malignancy (p=0.023) and preoperative bleeding (p=0.021) findings were observed more frequently in patients who underwent ERCP and went to surgery directly. It was observed that the stone size (p=0.004) and common bile duct diameter (p=0.038) values were higher in patients who underwent ERCP and went directly to surgery. Among the patients who underwent stent+UDCA, three (4.9%) patients had perioperative bleeding, one (1.6%) patient had preoperative perforation, four (6.6%) had postoperative pancreatitis, and one (1.6%) had mortality. The mean hospital stay was 1.96±2.1 days (**Table 1**). The clinical and demographic data of the patients who received a temporary plastic stent after ERCP and were started on UDCA treatment are presented in **Table 2**.

Table 1. Clinical and demographic data of patients with difficult bile duct stones

	Stent + UDCA (n=61) n (%)	ERCP + Surgery (n=12) n (%)	pa
Gender			0.314
Female	30 (49.2)	4 (33.3)	
Male	31 (50.8)	8 (66.7)	
Heart disease	38 (62.3)	7 (58.3)	0.796
Kidney disease	8 (13.1)	2 (16.7)	0.744
Lung disease	8 (13.1)	2 (16.7)	0.744
Central nervous system disease	4 (6.6)	1 (8.3)	0.824
History of malignancy	-	1 (8.3)	0.023*
Pre-op bleeding	3 (4.9)	3 (25.0)	0.021*
Pre-op perforation	1 (1.6)	-	0.655
Post-op bleeding	-	-	-
Post-op pancreatitis	4 (6.6)	-	0.362
Mortality	1 (1.6)	-	0.655
	Stent + UDCA (n=61) Median	**ERCP + Surgery (n=12) Median**	**pb**
Age (year)	67.0	67	0.841
Stone Size (mm)	9.8	16	0.004**
Bile duct diameter (mm)	14	16	0.038*
Pre-op bilirubin (mg/dL)	0.9	2.25	0.512
Pre-op direct bilirubin (mg/dL)	0.4	1.55	0.627
Post-op bilirubin (mg/dl)	0.70	2.6	0.255
Post-op direct bilirubin (mg/dl)	0.30	1.75	0.277
Pre-op amylase (IU/L)	45.0	52	0.404
Post-op amylase (IU/L)	47.0	45	0.623
Duration of procedure (min)	15	27.5	0.365
Leukocyte (ų/L)	7.4	8.25	0.376
Length of stay (day)	1	1	0.825

* p<0.05, **p<0.001, a: chi-square and fisher exact test, b: Mann Whitney U test

Table 2. Clinical and demographic data of patients with stent insertion and UDCA treatment started for difficult bile duct stones

	Frequency (n)	Percentage (%)
Gender		
Female	30	49.2
Male	31	50.8
Heart disease	38	62.3
Kidney disease	8	13.1
Lung disease	8	13.1
Central nervous system disease	4	6.6
History of malignancy	-	-
	Mean±sd	**Median**
Age (year)	64.5±17.0	67.0
Stone size (mm)	9.5±2.4	9.8
Bile duct diameter (mm)	13.8±4.3	14
Pre-op bilirubin (mg/dl)	2.35±3.2	0.9
Pre-op direct bilirubin (mg/dl)	1.86±2.9	0.4
Post-op bilirubin (mg/dl)	1.81±2.7	0.70
Post-op direct bilirubin (mg/dl)	1.44±2.5	0.30
Pre-op amylase (IU/L)	107±389.3	45.0
Post-op amylase (IU/L)	124.8±281.5	47.0
Duration of procedure (min)	20.3±11.4	15
Leukocyte (ų/L)	8.3±5.4	7.4
Number of procedure (times)	2.2±0.5	2

sd: Standard deviation

Of the 61 patients with temporary plastic stent placement and UDCA treatment, 50 (81.96%) underwent two procedures, eight (13.11%) three procedures, and three (6.55%) four procedures. It was observed that the post-procedure total bilirubin ($p<0.001$) and direct bilirubin ($p<0.001$) values of the patients who underwent temporary plastic stent+UDCA were lower than before the procedure. There was no significant difference between the pre- and post- ERCP procedure findings of amylase value ($p>0.05$) (n=61). While a decrease was observed in total bilirubin and direct bilirubin values after all ERCP procedures, it was determined that the post-procedure total bilirubin values were lower in patients who had two procedures ($p=0.018$). On the other hand, direct bilirubin values were lower after the procedure in patients who had three procedures ($p=0.028$). The reduction in common bile duct diameter and stone size was found to be statistically significant in patients who underwent two procedures (respectively, $p<0.001$ and $p<0.001$). Although the reduction in stone size was statistically significant in those who underwent three procedures, the decrease in the diameter of the common bile duct was not significant (respectively, $p=0.011$ and $p=0.090$). In addition, although there was a numerical decrease in those who underwent four procedures, no statistically significant finding was observed (**Table 3**).

Table 3. Change in laboratory data in patients with stent + UDCA (n=61)

	Pre-operative Mean ± sd	Post-operative Mean ± sd	pa
Two procedures (n=50)			
Total bilirubin (mg/dL)	2.5±3.5	1.9±2.9	0.018*
Direct bilirubin (mg/dL)	1.9±3.2	1.6±2.6	0.066
Amylase (IU/L)	119.4±429.6	116.9±267.3	0.735
Bile duct diameter (mm)	16.5±3.4	13.6±4.2	<0.001**
Stone size (mm)	16.5±2.0	9.9±2.1	<0.001**
Three procedures (n=8)			
Total bilirubin (mg/dL)	2.4±1.8	1.4±1.0	0.063
Direct bilirubin (mg/dL)	1.9±1.9	1.0±1.1	0.028*
Amylase (IU/L)	47.6±26.5	204.1±415.6	0.237
Bile duct diameter (mm)	17.6±4.0	15.3±5.1	0.090
Stone size (mm)	16.4±3.0	7.3±2.9	0.011*
Four procedures (n=3)			
Total bilirubin (mg/dL)	1.9±1.6	0.6±0.6	0.180
Direct bilirubin (mg/dL)	1.5±1.6	0.15±0.07	0.180
Amylase (IU/L)	47±15.6	45.5±2.1	0.655
Bile duct Diameter (mm)	16.7±1.2	13.7±3.2	0.109
Stone size (mm)	16.8±1.3	7.4±2.8	0.109

* p<0,05, **p<0,001, a: Wilcoxon ranks test, sd: Standard deviation

DISCUSSION

Endoscopic retrograde cholangiopancreatography (ERCP) is an interventional method used in the diagnosis and treatment of pancreaticobiliary system pathologies. It has been used for many years in the treatment of common bile duct stones. With basic ERCP techniques that can be applied in many centers such as EST, ML, and EPBD, 85-90% of common bile duct stones can be treated. Bile duct stones that cannot be treated with these methods are called difficult bile duct stones and advanced ERCP procedures are needed. When these techniques are not available, surgical intervention or temporary biliary stenting techniques can be used (3,8,9,11).

In our study, the patients who underwent ERCP due to bile duct stones between the specified dates were examined and the clinical experience of temporary plastic stent application and UDCA treatment in patients who were accepted as having difficult bile duct stones was shared. While the first ERCP procedure was successful in 450 (84.9%) of 530 patients who were treated for bile duct stones, additional interventions were needed in 80 patients (15.1%) due to difficult bile duct stones. Our rate of detection of difficult bile duct stones was found to be compatible with the literature (3). Endoscopic sphincterotomy (EST), ML, and EPBD techniques can be used alone or in combination in the treatment of common bile duct stones. In a meta-analysis that compared EST and EPBD treatment, Park et al.(12) showed that stone clearance rates in the same session were better than EPBD in patients who underwent EST (EPBD vs. ES 0.59 odds ratio (OR) [0.36–0.94, 95% confidence interval (CI)]. There was no difference between EST and EST+EPBD (1.71 vs. 1.70 OR [0.92–3.17, 95% CI]). In another study involving 58 patients in whom EST was unsuccessful, EPBD was performed in addition to EST, and the procedure was successfully completed in 54 patients (93.1%). The remaining four patients (6.9%) needed ML (13). In cases where classical ERCP methods were not successful, advanced ERCP techniques were applied. In a study involving 44 patients with difficult bile duct stones, fully covered self-expandable stents (FCSEM) were used and became successful in 82% (14). In another study in which the efficacy and safety of EPLBD and EST were compared, Kogure et al. (15) demonstrated that EPLBD alone provided high success (EPLBD vs EST 90.7% vs. 78.8% $p=0.04$). In a study in which LL was used and 17 patients were treated, bile duct stones were cleared in the first session with a success rate of 94%. In another study involving 31 multicenter patients with LL, a success rate of 87% was achieved in the first session (16,17).

Temporary plastic stents can be used as another treatment method for difficult bile duct stones. This method can be used as a treatment method, as well as a bridge treatment until other interventions. Kedia et al. (18) used the multiple

plastic stent method in the management of difficult bile duct stones and showed that the stone size decreased and disappeared in some patients. A comparison was made between the patients with temporary plastic stent implantation and three-month replacement and patients in whom optional stent replacement was performed after stent insertion. Accordingly, the stone clearance rate was found to be higher in patients with frequent stent replacement. The rates of stent-related cholangitis were also found to be lower in the same group (19). It was shown that the addition of UDCA to temporary plastic stents leads to a reduction in stone size and ease of operation during stone removal (20). Similarly, UDCA treatment after stenting of the common bile duct was shown to be more effective than stenting alone (21). In another study conducted in our country, periodic stenting treatment was applied and a significant reduction in stone size was noted (22).

In our study, while the decrease in common bile duct diameter and stone size was statistically significant in patients who underwent two procedures, only the decrease in stone size was found to be significant in patients who underwent three procedures. Although there was a decrease in both stone size and diameter in patients who underwent four procedures, it was not statistically significant. This is thought to be due to the relatively low number of patients who underwent four procedures. However, Katsinelos et al. (23) emphasized that UDCA application after stenting did not significantly decrease the stone size in difficult common bile duct stones. Although there are different opinions in the literature, the general opinion is that stenting provides a clinical improvement in the disintegration of common bile duct stones by providing bile flow. It is not entirely clear how biliary stents can aid stone removal, but it is assumed that when the stents are left for a period of time, they cause mechanical friction against the stone and can lead to stone fragmentation, making it easier to clear in the subsequent ERCP. In addition, UDCA is known to dissolve bile duct stones by reducing intestinal cholesterol absorption and secretion of cholesterol into the bile (8,24,25). In the literature, success rates in the secondary ERCP procedure are given between 44-92%. It is reported that stone size is directly related to success (20). Similarly, in our study, successful results were obtained with two ERCP procedures in the majority of patients (81.96%). Successful stone extraction was performed in all patients, and mortality was observed in only one (1.6%) patient. There was a significant decrease in total and direct bilirubin values between the two ERCP procedures. Stone size and common bile duct diameter were found to be significantly higher in the group that went directly to surgery. This supports the literature in terms of emphasizing the importance of stone size. Our complication rates, such as bleeding and perforation, were found to be lower compared to the literature, while the length of hospital stay was consistent with the literature (25,26).

The main limitation of our study is that it was retrospective and could not be randomized with other ERCP techniques. The number of patients and the inability to make cost analysis can be described as other deficiencies.

CONCLUSION

In our study, temporary plastic stent application and 750mg/day UDCA treatment were shown to be beneficial in common bile duct stones that could not be removed with basic ERCP techniques in the first ERCP session. In the absence of any complications or emergency intervention, ERCP was performed at three-month intervals and common bile duct stones were successfully removed in all patients. On the other hand, our study is important because of being one of the few studies conducted on the issue in our country. In addition, considering the cost of advanced ERCP techniques, the lack of necessary equipment in every center, and the inaccessibility of patients to these applications, we think this practice is highly important.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was initiated with the approval of the Bandırma Onyedi Eylül University Faculty of Medicine Clinical Researches Ethics Committee (Date: 09.05.2022, Decision No: 2022-67).

Informed Consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

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Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

REFERENCES

1. Freitas ML, Bell RL, Dufy AJ. Choledocholithiasis: evolving standards for diagnosis and management. World J Gastroenterol 2006; 12: 3162–7.
2. Çetinkaya HB, Çay F. Can NLR be Used in discriminating simple and severe cholecystitis in emergency department admission?. Van Med J 2021; 28: 502-6.
3. Manes G, Paspatis G, Aabakken L, et al. Endoscopic management of common bile duct stones: European Society of Gastrointestinal Endoscopy (ESGE) guideline. Endoscopy 2019; 51: 472-91.
4. Anderloni A. Difficult common bile duct stones: still "difficult" or just "different"?. Endoscopy 2020; 52: 429-30.
5. Yang J, Peng JY, Chen W. Endoscopic biliary stenting for irretrievable common bile duct stones: Indications, advantages, disadvantages, and follow-up results. Surgeon 2012; 10: 211–7.

6. Ye X, Huai J, Sun X. Efectiveness and safety of biliary stenting in the management of difficult common bile duct stones in elderly patients. Turk J Gastroenterol 2016; 27: 30–6.
7. Lee TH, Han JH, Kim HJ, et al. Is the addition of choleretic agents in multiple double-pigtail biliary stents effective for difficult common bile duct stones in elderly patients? A prospective, multicenter study. Gastrointest Endosc 2011; 74: 96–102.
8. Slattery E, Kale V, Anwar W, Courtney G, Aftab AR. Role of long-term biliary stenting in choledocholithiasis. Digestive Endoscopy 2013; 25: 440-3.
9. Hormati A, Ghadir M, Sarkeshikian SS, Modarres MP, Rafiei M, Alemi F. Efficacy of common bile duct stenting for large stones. Govaresh 2017; 22: 149-53.
10. Yamamoto R, Tazuma S, Kanno K, et al. Ursodeoxycholic acid after bile duct stone removal and risk factors for recurrence: a randomized trial. J Hepatobiliary Pancreat Surg 2016; 23: 132-6.
11. Johnson KD, Perisetti A, Tharian B, et al. Endoscopic retrograde cholangiopancreatography-related complications and their management strategies: a "scoping" literature review. Dig Dis Sci 2020; 65: 361-75.
12. Park CH, Jung JH, Nam E, et al. Comparative efficacy of various endoscopic techniques for the treatment of common bile duct stones: a network meta-analysis. Gastrointest Endosc 2018; 87: 43-57.
13. Ersoz G, Tekesin O, Ozutemiz AO, Gunsar F. Biliary sphincterotomy plus dilation with a large balloon for bile duct stones that are difficult to extract. Gastrointest Endosc 2003; 57: 156-9.
14. Hartery K, Lee CS, Doherty GA, et al. Covered self-expanding metal stents for the management of common bile duct stones. Gastrointest Endosc 2017; 85: 181-6.
15. Kogure H, Kawahata S, Mukai T, et al. Multicenter randomized trial of endoscopic papillary large balloon dilation without sphincterotomy versus endoscopic sphincterotomy for removal of bile duct stones: Marvelous trial. Endoscopy 2020; 52: 736-44.
16. Wong JC, Wong MY, Lam KL, Lau JY. Second-generation peroral cholangioscopy and holmium: YAG laser lithotripsy for rescue of impacted biliary stone extraction basket. Gastrointest Endosc 2016; 83: 837-8.
17. Navaneethan U, Hasan MK, Kommaraju K, et al. Digital, single-operator cholangiopancreatoscopy in the diagnosis and management of pancreatobiliary disorders: a multicenter clinical experience (with video). Gastrointest Endosc 2016; 84: 649-55.
18. Kedia P, & Tarnasky PR. Endoscopic management of complex biliary stone disease. Gastrointest Endosc Clinics 2019; 29: 257-75.
19. Di Giorgio P, Manes G, Grimaldi E, et al. Endoscopic plastic stenting for bile duct stones: stent changing on demand or every 3 months. A prospective comparison study. Endoscopy 2013; 45: 1014-7.
20. Han J, Moon JH, Koo HC, et al. Effect of biliary stenting combined with ursodeoxycholic acid and terpene treatment on retained common bile duct stones in elderly patients: a multicenter study. ACG 2009; 104: 2418-21.
21. Hormati A, Ghadir MR, Sarkeshikian SS, et al. Adding ursodeoxycholic acid to the endoscopic treatment and common bile duct stenting for large and multiple biliary stones: Will it improve the outcomes?. BMC Gastroenterol 2020; 20: 1-5.
22. Ödemiş B, Kuzu UB, Öztaş E, et al. Endoscopic management of the difficult bile duct stones: a single tertiary center experience. Gastroenterol Res Pract 2016.
23. Katsinelos P, Kountouras J, Paroutoglou G, Chatzimavroudis G, Zavos C. Combination of endoprostheses and oral ursodeoxycholic acid or placebo in the treatment of difficult to extract common bile duct stones. Dig Liver Dis 2008; 40: 453-9.
24. Slattery E, Kale V, Anwar W, Courtney G, Aftab AR. Role of long-term biliary stenting in choledocholithiasis. Digestive Endoscopy 2013; 25: 440-3.
25. Hofmann AF. Medical treatment of cholesterol gallstones by bile desaturating agents. Hepatology 1984; 4: 199-208.
26. Banerjee N, Hilden K, Baron TH, Adler DG. Endoscopic biliary sphincterotomy is not required for transpapillary SEMS placement for biliary obstruction. Dig Dis Sci 2011; 56: 591-5.
27. Çay F, Girgin M. Is laparoscopic cholecystectomy safe in acute cholecystitis?. Health Sci J Adıyaman Universty 2017; 3: 487-95.

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Systemic immune inflammation index: is it a new marker for contrast-induced nephropathy?

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ABSTRACT

Introduction: Worldwide, >200 million patients are affected by peripheral arterial disease (PAD) and endovascular interventional treatments are increasingly being applied. Contrast-induced nephropathy (CIN) is the third most common cause of renal failure in hospitals. However, factors such as renal vasoconstriction, decrease in renal blood flow, endothelial dysfunction, and oxidative stress have been suggested in the etiology of CIN. Studies are showing that inflammatory markers increase in CIN. Systemic immune inflammation index (SII), a newly defined parameter, is calculated by multiplying the platelet and lymphocyte counts and dividing by the neutrophil count. Studies are showing that this parameter influences prognosis in various cancer types. Considering that inflammation may play a role in CIN, we planned this study to investigate the role of SII in patients undergoing percutaneous peripheral vascular interventions.

Material and Method: 300 patients who underwent percutaneous peripheral vascular interventions between August 2018-December 2021 due to peripheral arterial disease were included in the study. The data of the patients were scanned retrospectively from the patient files. The neutrophil-lymphocyte ratio (NLR) was calculated by dividing the neutrophil count by the lymphocyte count. SII was found by multiplying NLR with platelet count

Results: Contrast-induced nephropathy developed in 41 (12.3%) patients. CIN(+) patients also, had higher CRP levels (5.1±0.7vs 2.4±0.4,P<0.05), NLR (4.07±1.07vs 2.65±0.84, P<.005), SII score (1778±627.57vs 867.14±491.88, P<.005.) the contrast media used was also higher in CIN(+) patients (176.19±48.44 vs 128.72±48.44;P<0.05) Multivariate logistic regression analysis demonstrated that a high SII score was an independent predictor of development of CIN (odds ratio [OR]: 1.002, 95% confidence interval [CI]: 1.001-1.002, P<.0005) together with high NLR (OR: 3.56, 95% CI: 1.905-6.675, P<.005) and CRP (OR: 1.002, 95% CI: 1.001-1.002, P<.005 Receiver operating characteristic curve analysis demonstrated that the best cutoff value of 1224 for SII to predict the development of CIN with 85% sensitivity and 72% specificity (area under ROC curve 0.904 [95% CI: 0.866-0.942], P<.005).

Conclusion: Imbalance in inflammatory cells, the increase in neutrophils, and the decrease in lymphocytes play a role in developing kidney damage. Impaired immune functions due to lymphocytopenia contribute to the development of acute kidney injury. Oxidative stress exacerbates the inflammatory state by increasing inflammatory cell infiltration. AS a result, SII may be a powerful predictor of inflammation and can be used to determine the risk before interventional procedures.

Keywords: contrast-induced nephropathy, inflammation, peripheral vascular interventions, systemic immune inflammation index

INTRODUCTION

Worldwide, >200 million patients are affected by the peripheral arterial disease (PAD and endovascular interventional treatments are increasingly being applied (1). Contrast-induced nephropathy (CIN) is hospitals' third most common cause of renal failure (2). It is usually reversible but related to increased morbidity and mortality (3).

Although factors such as renal vasoconstriction, decrease in renal blood flow, endothelial dysfunction, and oxidative stress have been suggested in the etiology of CIN, it is not known why some patients with the same risk factors develop nephropathy while others do not (4,5). Studies show that inflammatory markers increase in CIN (6-10).

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Systemic immune inflammation index (SII), a newly defined parameter, is calculated by multiplying the platelet and lymphocyte counts and dividing by the neutrophil count. Studies show that this parameter influences prognosis in various cancer types (11-14).

Considering that inflammation may play a role in CIN, we planned this study to investigate the role of SII in patients undergoing percutaneous peripheral vascular interventions.

MATERIAL AND METHOD

The study was initiated with the approval of the Prof. Dr. Cemil Taşçıoğlu City Hospital Institutional Ethical Committee (Date: 2022, Decision No: E-48670771-59.99). All procedures were carried out under the ethical rules and the principles of the Declaration of Helsinki.

Patients who underwent percutaneous peripheral vascular interventions between August 2018-December 2021 due to peripheral arterial disease were included in the study. The data of the patients were scanned retrospectively from the patient files. Risk factors such as age, gender, diabetes mellitus (DM), hypertension (HT), dyslipidemia, smoking, and drugs were scanned from patient files. Exclusion criteria were the presence of active infection, presence of chronic inflammatory or autoimmune disease, known cancer history, presence of chronic liver disease, end-stage renal failure (GFR<10 ml/min), and heart failure (EF<40).

All laboratory parameters before the procedure and three days after the procedure were recorded from the files. CIN was defined as a 25% or >0.5 mg/dl increase in creatinine value on the third day (15).

The neutrophil-lymphocyte ratio (NLR) was calculated by dividing the neutrophil count by the lymphocyte count. SII was found by multiplying NLR with platelet count.

SPSS (Statistical Package for Social Sciences, Chicago, IL) for Windows 20.0 was used for statistical analysis. Data about continuous variables were expressed in mean±standard deviation if otherwise is not indicated. Intergroup comparisons were made with Student's t-test (in data with a normal distribution) or with Mann–Whitney U test (in data without a normal distribution). Categorical variables were compared with the Chi-square test. Pearson's correlation coefficient was used for continuous variables with normal distribution, and Spearman's correlation coefficient was used for continuous variables that are not normally distributed p<0.05 was considered significant. The effects of different variables on the development of CIN were calculated with univariate analysis. The model included parameters with a P<.10 in univariate analysis for multivariate regression analysis. The cutoff level of SII and NLR in predicting CIN formation was determined by performing a receiver operating characteristic curve (ROC) analysis. The value corresponding to the highest sensitivity and specificity value in the ROC analysis was accepted as the optimal cutoff value. A 2-sided P<.05 was considered significant.

RESULTS

Of the 300 patients included in the study, 228 (76.3%) were male. The mean age was 62±12.3 years. One hundred sixty patients had diabetes (53.3%), 105 had hypertension (35%), and 202 were smokers (67.3%). The mean creatinine level was 1.06±0.75mg/dl, and CRP was 5.25±1.92. Laboratory findings of the patients can be seen in **Table 1**.

Table 1. Demographic properties of study population

Age (years)	62±12.18
Male (n,%)	228 (76.3%)
Diabetes mellitus (n,%)	160 (53.3%)
Hypertension (n,%)	105 (35%)
Smoking (n,%)	202 (67.3%)
Glucose (mg/dL)	154.3±79.61
Creatinine (mg/dL)	1.06±0.75
AST (U/L)	24.52±12.18
ALT (U/L)	20.24±10.75
T. Cholesterol (mg/dL)	196.29±52.79
HDL (mg/dL)	43.05±10.52
LDL (mg/dL)	136.42±64.37
Triglyceride (mg/dL)	165.99±121.61
Hgb (g/L)	13.65±2.22
Albumin (g/dL)	3.87±0.5
Contrast volume	135.0±40.6
CRP (mg/L)	5.25±1.92
NLR	2.84±1.01
SII	994.76±40.65

Contrast-induced nephropathy developed in 41 (12.3%) patients. the patients with and without CIN are summarized in Table 1. The age of the patients was similar between the two groups. (62.04±11.5 in CIN (-) vs 61.7±15.73 years in CIN (+); P>0.05). The rate of patients with HT and DM was also similar between CIN (-) and CIN (+) (36.8% vs 23.8%, P>0.05; 51.6% vs 64.3%, P>.005, respectively). Other demographic characteristics and the previous medications were similar between both groups. There was no significant difference between the two groups regarding sex and smoking (**Table 2**).

When the hematological parameters were analyzed between the two groups, there was no significant difference in hemoglobin (P>.05).

There was no statistically significant difference between groups in terms of glucose, AST, ALT, total cholesterol, HDL, LDL and triglyceride levels (p>0.05, **Table 1**).

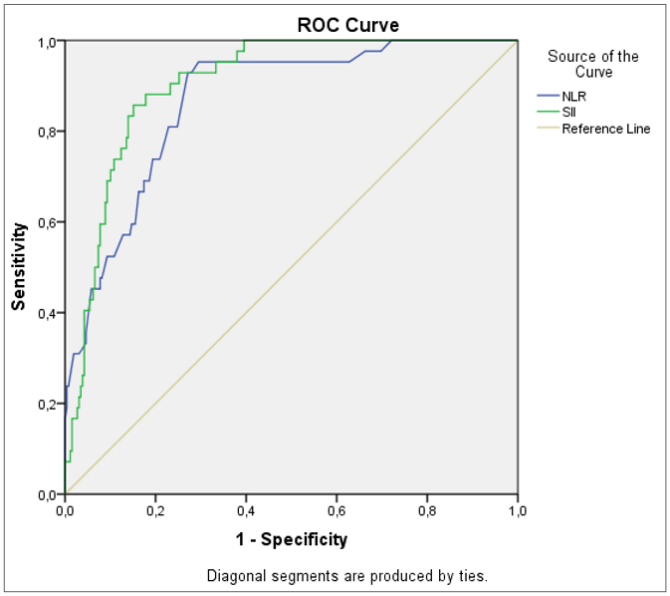
The patients who had CIN had statistically significantly higher creatinine levels (1.89±1.54 vs 0.92±0.17; p<0.05). CIN (+) patients also, had higher CRP levels (5.1±0.7vs 2.4±0.4, P<0.05), NLR (4.07±1.07vs 2.65±0.84, P<.005), SII score (1778±627.57vs 867.14±491.88, P<.005.) the contrast media used was also higher in CIN (+) patients (176.19±48.44 vs 128.72±48.44; P<0.05). CIN (+) patients had significantly higher T.Chol and LDL levels (214.54±57.75vs 193.6±51.65; p=0.043 and 150.19±60.3 vs 134.17±64.83; p_0.021).

Table 2. Comparision of the groups

	CIN(-)	CIN(+)	P
Age (years)	56.81±13.74	62.84±11.72	0.013
Male(n,%)	196 (76.3%)	32 (76.2%)	0.992
Diabetes Mellitus (n,%)	139 (53.9%)	21 (50%)	0.641
Hypertension(n,%)	88 (34%)	17 (40.5%)	0.422
Smoking (n,%)	170 (65.9%)	32 (76.2%)	0.187
Glucose (mg/dl)	154.43±78.38	153.48±87.77	0.493
Creatinine (mg/dl)	0.92±0.17	1.89±1.54	0.023
AST(U/L)	24.92±14.29	22.1±13.1	0.930
ALT(U/L)	20.14±14.72	20.83±10.84	0.357
T.Cholesterol(mg/dl)	193.6±51.65	214.54±57.75	0.043
HDL(mg/dl)	43.12±10.75	42.62±9.03	0.713
LDL(mg/dl)	134.17±64.83	150.19±60.39	0.021
Triglyceride (mg/dl)	164.91±122.42	171.9±117.77	0.954
Hgb (g/L)	13. ±1.02	13.14±2	0.590
Albumin(gr/dl)	3.85±0.55	3.96±0.5	0.810
CRP (mg/L)	2.4±0.4	5.1±0.	0.01
Contrast volume	128.72±48	176.19±48.4	0.001
NLR	2.65±0.84	4.07±1.07	0.00
SII	867.14±491.88	1778±627.57	0.00

The patients who had CIN were grouped in two according to gender. Thirty-two of the patients were male. DM, hypertension, and smoking incidence were similar (**Table 3**). There was no difference in terms of contrast volume, NLR, and SII (152±32 VS 117±55; P=0.125, 2.64±0.4 vs±2.62±0.89; P=0.257, 853.14±468.88 vs. 915.3±526; P=0.467)

The role of several CIN risk factors was also evaluated by multivariate analysis. This included age, gender, DM, HT, contrast volume, serum creatinine, glucose, NLR, CRP, and SII. Multivariate logistic regression analysis demonstrated that a high SII score was an independent predictor of development of CIN (odds ratio [OR]: 1.002, 95% confidence interval [CI]: 1.001-1.002, P<.0005) together with high NLR (OR: 3.56, 95% CI: 1.905-6.675, P<.005) and CRP (OR: 1.002, 95% CI: 1.001-1.002, P<.005. Age was not a significant indicator (OR: 1.000%95 CI: 0.928-1.001,P>0.05). Receiver operating characteristic curve analysis demonstrated that the best cutoff value of 1224 for SII to predict the development of CIN with 85% sensitivity and 72% specificity (area under ROC curve 0.904 [95% CI: 0.866-0.942], P<.005). For NLR, the best cutoff value of 3.17 predicted the development of CIN with a sensitivity of 92% and specificity of 72%, and the area under the curve was 0.867 (95% CI: 0.814-0.919; P<.005; **Figure 1**).



DISCUSSION

The most important finding of this study is that the increase in SII score, a new inflammation parameter, is a robust independent predictor of CIN.

According to previous studies, while the incidence of CIN is below 2% in patients without risk factors, the incidence rises to 90% in patients with risk factors (6). The incidence of CIN was 12.3% in our study. This rate corresponded with the incidence of CIN in patients who underwent angiography for the acute coronary syndrome (16).

In our study, basal creatinine of the CIN (+) group was statistically significantly higher. Preexisting chronic kidney disease is a known risk factor for contrast-induced nephropathy. In a series of 1144 patients, Davidson et al. l. investigated patients undergoing cardiac catheterization and documented a low risk of CIN (increment of creatinine levels of at least 0.5 mg/dL) in patients with normal renal function compared to those with preexisting CKD (creatinine levels exceeding 1.2 mg/dL). These investigators found that the risk for CIN increased significantly (20%) when serum creatinine exceeded 2.0 mg/dL (17).

The age of the CIN (+) group was statistically significantly higher in our study. This is concordant with other studies (18, 19). This is possibly caused by the decline in renal function with increasing age. Vascular stiffens are increased, and vasodilator response is decreased by aging. Also, pluripotent stem cells decreased in advanced age, causing a decrease in vascular repair (20).

Contrast volume is increased in CIN (+) group. High doses and repeated use of contrast material administered within 72 hours increase CIN (+). This is more common in the first-generation contrast agents (21, 22)

In the CIN (+) group, T.Chol. and LDL levels were increased compared with the CIN (-) group. This is consistent with the literature. According to statin use, in a study by Hoshi et al. (23), 2198 patients were analyzed. In the statin pretreatment group, CIN was observed less. Statin may reduce contrast-induced inflammation and may have beneficial effects against CIN. Hyperlipidemia may increase systemic inflammation and disturb tubular function (24).

A high SII score indicates decreased immune system with an increased inflammatory state. To understand the relationship between the SII index and CIN, the roles of neutrophils, platelets, and lymphocytes should be evaluated separately.

Inflammation is a pre-thrombotic condition (25). Endothelial damage caused by inflammation leads to a pre-thrombotic state. In addition, inflammatory cells reduce the amount of critical anticoagulant substances (26)

In this inflammatory process, platelets are activated by chemokines, secreted proteins, and microRNAs (27). Activation of the coagulation system and downregulation of the anticoagulant system causes an increase in platelet levels and an increased risk of CIN (27). Experimental studies have shown that the imbalance in inflammatory cells, the increase in neutrophils, and the decrease in lymphocytes play a role in developing kidney damage (28), stimulation of neutrophils increases vascular permeability, and endothelial damage occurs (29, 30).

Impaired immune functions due to lymphocytopenia contribute to the development of acute kidney injury (30). Oxidative stress exacerbates the inflammatory state by increasing inflammatory cell infiltration (31). Detection of the inflammatory process can be used to determine the risk before interventional procedures.

SII is thought to show inflammatory processes better than NLR and platelet-lymphocyte ratio. This is based on the findings in recent studies showing the relationship of SII with poor outcomes in various diseases (11-13, 32-35).

Xu et al. (36) showed that SII is associated with acute kidney injury in patients with hepatocellular carcinoma who underwent hepatectomy. Bağcı et al. (37) showed that it is an independent marker of CIN in patients with myocardial infarction. Yang et al. (32) claimed that the increase in SII scores was superior to traditional risk factors in predicting mortality and morbidity in coronary arterial disease. Gok et al. (38). Reported a relationship between high SII scores and the severity of pulmonary embolism. Our study found that among the inflammatory markers we examined in peripheral angiography patients, the SII was the most decisive and independent marker associated with CIN development. Moreover, we identified that the optimum cutoff point for the SII was 1224, which predicted the risk of developing CIN with 85% sensitivity and 72% specificity.

This study has some limitations. Few patients were included in the study, and the data were reviewed retrospectively. The study was a single-center study. SII levels were calculated at admission. Control creatinine level was measured three days after contrast use; therefore, it could not be detected if there was an increase after 72 hours.

Table 3. Comparision of the CIN(+) group in terms of gender

	Male (n=32)	Female (n=10)	P
Age (years)	63.35±9.54	61.18±11.72	0.4
Diabetes Mellitus (n,%)	17 (53.1%)	4 (40.3%)	0.641
Hypertension(n,%)	11 (34.4%)	4 (40%)	0.422
Smoking (n,%)	28 (87.5%)	32 (76.2%)	0.182
Glucose (mg/dl)	157.24±81.88	145.57±66.55	0.5
Creatinine (mg/dl)	1.1±0.72	0.93±1.5	0.12
AST(U/L)	24.92±4.29	32.97±6.46	0.881
ALT(U/L)	18.52±12.61	25.64±4.3	0.343
T.Cholesterol (mg/dl)	193.6±51.65	214.54±57.75	0.521
HDL (mg/dl)	43.12±10.75	42.62±9.03	0.513
LDL (mg/dl)	135.3±68.25	130.54±53.2	0.413
Triglyceride (mg/dl)	172.94±133.1	139.08±73.22	0.954
Hgb (g/L)	13.82. ±1.02	13.55±2	0.51
Albumin (g/dl)	3.85±0.55	3.78±0.5	0.243
CRP (mg/L)	2.76±0.4	3.48±0.	0.112
Contrast volume	152±32	117±55	0.125
NLR	2.64±0.83	2.62±0.89	0.257
SII	853.14±468.88	915.3±526	0467

Table 4. Binary logistic regression analysis

	Odds ratio	Cl95%	P
Age	1.000	0.928-1.001	0.057
CRP	1.002	1.001-1.002	0.003
NLR	3.566	1.905-6.675	0.000
SII	1.002	1.001-1.002	0.000

CONCLUSION

This study determined that high SII was an independent indicator of the development of CIN in patients who underwent percutaneous peripheral vascular interventions.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was initiated with the approval of the Prof. Dr. Cemil Taşçıoğlu City Hospital Institutional Ethical Committee (Date: 2022, Decision No: E-48670771-59.99).

Informed Consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors declare no conflicts of interest.

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REFERENCES

1. Song P, Rudan D, Zhu Y, et al. Global, regional, and national prevalence and risk factors for peripheral artery disease in 2015: an updated systematic review and analysis. Lancet Glob Health 2019; 7: 1020–30.
2. Pucelikova T, Dangas G, Mehran R. Contrast-induced nephropathy. Catheter Cardiovasc Interv 2008; 71: 62-72.
3. Morcos R, Kucharik M, Bansal P, et al. Contrast-induced acute kidney injury: review and practical update. Clin Med Insights Cardiol 2019; 13: 1179546819878680.
4. James MT, Samuel SM, Manning MA, et al. Contrast-induce acute kidney injury and risk of adverse clinical outcomes after coronary angiography: a systematic review and meta-analysis. Circ Cardiovasc Interv 2013; 6: 37-43.
5. Caiazza A, Russo L, Sabbatini M, Russo D. Hemodynamic and tubular changes induced by contrast media. Biomed Res Int 2014; 2014: 578974.
6. Goldenberg I, Matetzky S. Nephropathy induced by contrast media: pathogenesis, risk factors and preventive strategies. CMAJ 2005; 172: 1461-71.
7. Kurtul A, Yarlioglues M, Duran M, Murat SN. Association of neutrophil-to lymphocyte ratio with contrast-induced nephropathy in patients with non-st-elevation acute coronary syndrome treated with percutaneous coronary intervention. Heart Lung Circ 2016; 25: 683-90.
8. Kaya A, Kaya Y, Topcu S, et al. Neutrophil-to-lymphocyte ratio predicts contrast-induced nephropathy in patients undergoing primary percutaneous coronary intervention. Angiology 2014; 65: 51-6.
9. Liu Y, Tan N, Zhou YL, et al. High-sensitivity C-reactive protein predicts contrast-induced nephropathy after primary percutaneous coronary intervention. J Nephrol 2012; 25: 332-40.
10. Kocas C, Yildiz A, Abaci O, et al. Platelet- to-lymphocyte ratio predicts contrast-induced nephropathy in patients with non-st segment elevation acute coronary syndrome. Angiology 2015; 66: 964-8.
11. .Ji Y, Wang H. Prognostic prediction of systemic immune inflammation index for patients with gynecological and breast cancers: a meta-analysis. World J Surg Oncol 2020; 18: 197.
12. Huang Y, Gao Y, Wu Y, Lin H. Prognostic value of systemic immune-inflammation index in patients with urologic cancers: a meta-analysis. Cancer Cell Int 2020; 12: 499
13. Zhang Y, Lin S, Yang X, Wang R, Luo L. Prognostic value of pretreatment systemic immune-inflammation index in patients with gastrointestinal cancers. J Cell Physiol 2019; 234: 5555-6
14. Kelesoglu S, Yilmaz Y, Elcık D, et al. Systemic immune inflammation index: a novel predictor of contrast-induced nephropathy in patients with non-st segment elevation myocardial infarction. Angiology 2021 Oct; 72: 889-95.
15. Mehran R, Aymong ED, Nikolsky E, et al. A simple risk score for prediction of contrast-induced nephropathy after percutaneous coronary intervention: development and initial validation. J Am Coll Cardiol 2004; 44: 1393-9.
16. Tsai TT, Patel UD, Chang TI, et al. Contemporary incidence, predictors, and outcomes of acute kidney injury in patients undergoing percutaneous coronary interventions: insights from the NCDR Cath-PCI registry. JACC Cardiovasc Interv 2014; 7: 1-9.
17. Davidson CJ, Hlatky M, Morris KG, et al. Cardiovascular and renal toxicity of a nonionic radiographic contrast agent after cardiac catheterization. A prospective trial. AnnIntern Med 1989; 110: 119-24
18. Eng J, Wilson RF, Subramaniam RM, et al. Comparative effect of contrast media type on the incidence of contrastinduced nephropathy: a systematic review and meta-analysis. Ann Intern Med 2016; 164: 417–24.
19. Wong PC, Li Z, Guo J, Zhang A. Pathophysiology of contrast-induced nephropathy. Int J Cardiol 2012; 158: 186– 92.
20. Brandes RP, Fleming I, Busse R. Endothelial aging. Cardiovasc Res 2005; 66: 286 94.
21. Jurado-Román A, Hernández-Hernández F, García-Tejada J, et al. Role of hydration in contrast-induced nephropathy in patients who underwent primary percutaneous coronary intervention. Am J Cardiol 2015; 115: 1174-8.
22. Morcos SK, Thomsen HS, Webb JA. Contrast-media-induced nephrotoxicity: A consensus report. Contrast media safety committee, European Society of Urogenital Radiology (ESUR). Eur Radiol 1999; 9: 1602-13.
23. Hoshi T, Sato A, Kakefuda Y, et al. Preventive effect of statin pretreatment on contrast-induced acute kidney injury in patients undergoing coronary angioplasty: propensity score analysis from a multicenter registry. Int J Cardiol 2014; 171: 243-9.
24. Kwasa EA, Vinayak S, Armstrong R. The role of inflammation in contrast-induced nephropathy. Br J Radiol 2014; 87; 20130738.
25. Lau A, Chung H, Komada T, et al. Renal immune surveillance and dipeptidase-1 contribute to contrast-induced acute kidney injury. J Clin Invest 2018; 128: 2894–913.
26. Lu Z, Cheng D, Yin Y, et al. Antithrombin iii protects against contrast-induced nephropathy, E Bio Medicine 2017; 17: 101–7.
27. Wang F, Yin J, Lu Z, et al. Limb ischemic preconditioning protects against contrast-induced nephropathy via renalase.E Bio Medicine 2016; 9: 356–65.
28. Sun SC. The non-canonical nf-κb pathway in immunity and inflammation, Nat Rev Immunol 2017; 17: 545–58.
29. Zhang Q, Lenardo MJ, Baltimore D. 30 years of nf-κb: a blossoming of relevance to human pathobiology. Cell 2017; 168: 37–57.
30. Machado RA, Constantino Lde S, Tomasi CD, et al. Sodium butyrate decreases the activation of nf-κb reducing inflammation and oxidative damage in the kidney of rats subjected to contrast-induced nephropathy. Nephrol Dial Transplant 2012; 27: 3136–40.
31. Furuichi K, Wada T, Iwata Y, et al. Interleukin-1- dependent sequential chemokine expression and inflammatory cell infiltration in ischemia-reperfusion injury. Crit Care Med 2006; 34: 2447–55.
32. Huang J, Zhang Q, Wang R, et al. Systemic immune-inflammatory index predicts clinical outcomes for elderly patients with acutemyocardial infarction receiving percutaneous coronary intervention. Med Sci Monit 2019; 25: 9690-701.
33. Yang YL, Wu CH, Hsu PF, et al. Systemic immune-inflammation index (SII) predicted clinical outcome in patients with coronary artery disease. Eur J Clin Invest 2020; 50: e13230.
34. Li B, Li W, Li X, Zhou H. Inflammation: A novel therapeutic target/direction in atherosclerosis. Curr Pharm Des 2017; 23: 1216-27.

35. Seo M, Yamada T, Morita T, et al. Prognostic value of systemic immune-inflammation index in patients with chronic heart failure. Eur Heart J 2018; 39: ehy564.P589.
36. Xu J, Hu S, Li S, et al. Systemic immuneinflammation index predicts postoperative acute kidney injury in hepatocellular carcinoma patients after hepatectomy. Medicine 2021; 100: e25335.
37. Bagci A, Aksoy F, Ba¸s HA. Systemic immune-inflammation index may predict the development of contrast-induced nephropathy in patients with ST-segment elevation myocardial infarction. Angiology 2022; 73: 218– 24.
38. Gok M, Kurtul A. A novel marker for predicting severity of acute pulmonary embolism: systemic immune-inflammation index. Scand Cardiovasc J 2021; 55: 91-6.

Original Article

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Effect of preoperative hypoalbuminemia on postoperative mortality and morbidity in liver transplant surgery

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ABSTRACT

Aim: In this study, it isaimed to retrospectively evaluatethe effect of serum albumin levels on postoperative mortality and morbidity in patients with end-stage liver failure with hypoalbuminemia who underwent follow-up liver transplant surgery.

Material and Method: After the approval of the ethics committee, the patients who underwent liver transplant surgery between April 2011 and September 2016 were divided into two groups as with preoperative albumin values <3.5 g/dl and ≥3.5 g/dl. Demographic characteristics of the patients, preoperative and postoperative arterial blood gases, posteroanterior chest radiographs, tracheal aspirate cultures, preoperative and postoperative serum creatinine levels, mechanical ventilation and intensive care unit length of stay, transfusion of blood and blood products, and early postoperative complications (30 days) were determined and compared retrospectively.

Result: It was observed that mechanical ventilation, intensive care hospitalization and discharge times were higher in the group with low albumin levels ($p<0.05$). More blood and blood products were used in the group with low albumin levels ($p<0.05$). Postoperative pulmonary complications and acute kidney injury were more common in the group with low albumin levels ($p<0.05$). Complications in the early postoperative period were more common in the group with low albumin levels.

Conclusion: As a result, it isconcluded that the preoperative serum albumin level can be used as a guide and a valuable parameter in the postoperative follow-up in the detection of perioperative and postoperative complications in liver transplant recipients.

Keywords: Hypoalbuminemia, liver transplantation, mortality, morbidity

INTRODUCTION

Hypoalbuminemia is a picture encountered in many acute or chronic clinical conditions. In 20% of hospitalizations, patients are hypoalbuminemia. Low albumin levels are accepted as an indicator of poor prognosis. However, it has been shown that preoperative hypoalbuminemia is a strong indicator of mortality and morbidity in the early postoperative period in patients undergoing cardiac surgery, gastrointestinal surgery, orthopedic surgery, sepsis and major infections (1).

Albumin is a plasma protein responsible for most of the plasma oncotic pressure and is also a negative acute phase reactant. Albumin levels are an informative parameter for the general condition of the patient, both as an acute phase reactant and as an indirect indicator of the patient's metabolic status and organ functions (such as liver). Patients with liver failure are frequently found to have low plasma albumin. It is characterized by impaired hepatocellular functions and a decrease in albumin synthesis up to 60-80% in advanced stages of cirrhosis (2). Albumin has found its place as a prognostic marker in various diseases. Albumin levels are frequently used for estimating the clinical course and survival of patients, since it is a relatively cheaper and accessible test compared to special tests, which are checked in routine biochemical tests.

Based on these effects of albumin, the present study was arrangedwith the hypothesis that serum albumin level may play a role in predicting postoperative morbidity and mortality in patients with end-stage chronic liver disease who are scheduled for liver transplantation with hypoalbuminemia.

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MATERIAL AND METHOD

The study was initiated with the approval of the Uludağ University Medical Faculty Clinical Researches Ethics Committee (Date: 2017, Decision No: 4/37). All procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki.

The study was conducted by retrospectively scanning the files of liver transplant recipients who were performed at Uludağ University Faculty of Medicine between April 2011 and September 2016. The obtained 136 files were examined. Patients over the age of 18, who underwent liver transplantation were included intothis study, while patients who were hospitalized in the intensive care unit in the preoperative period, patients under the age of 18 and those who died in the first 24 hours postoperatively were excluded. In this context, 9 patient files were excluded from the study, since 3 patients were under the age of 18, 4 patients were operated under preoperative intensive care treatment and 2 patients died within 24 hours postoperatively.

Age, gender and MELD (Model For End-stage Liver Disease) scores of the patients were recorded in order to determine the differences between the study groups of the patients' demographic characteristics and the severity of their current disease.

In order to determine the effects of serum albumin level on postoperative mortality and morbidity, 127 screened patient files were divided into two groups according to their preoperative serum albumin level.

Group I: patients with preoperative serum albumin level below 3.5 g/dL

Group II: patients with preoperative serum albumin level of 3.5gr/dL and above

In order to compare the respiratory functions and postoperative respiratory complications, posteroanterior (PA) chest X-ray and tracheal aspirate cultures were evaluated on postoperative day 1, and mechanical ventilation times were recorded.

Preoperative, postoperative 1st day and postoperative 2nd day creatinine levels of the patients were also recorded and the creatinine increase between the groups and acute kidney injury rates were tried to be compared by using the AKIN (Acute Kidney Injury Network) classification. (**Table 1**)

In addition, numbers of erythrocyte suspension, fresh frozen plasma and thrombocyte suspension transfused in the perioperativeperiod,the duration of intensive care unit stay and the period from the end of the operation to the discharge were documented.

Table 1. Classification of acute kidney injury AKIN

Stage	Serum creatinine	Urine output
1	0.3 mg/dl or 1.5-2 times increase in serum creatinine compared to baseline	<0.5 ml/kg per hour for 6 hours
2	2-3 times increase in serum creatinine compared to baseline	<0.5 ml/kg per hour for 12 hours
3	3-fold increase in serum creatinine compared to baseline or 0.5 mg/dl increase in creatinine with serum creatinine ≥4 mg/dl	12 hour anuria or <0.3 ml/kg/hour for 24 hours or renal replacement therapy

When the clinical course and epicrisis of the patients were examined, a table was created from the complications observed in the postoperative 30 days that would prolong the most common mortality, morbidity and hospital stay, and the complications observed in the clinical course of the patients were recorded.

Statistical Analysis

In thisstudy, mean, standard deviation, median, lowest, highest, frequency and ratio values were used in the descriptive statistics of the data. The distribution of variables was measured viaKolmogorov-Smirnov test. The Mann-Whitney U test was used for the analysis of quantitative independent data. Wilcoxon test was used for the analysis of dependent quantitative data. Fischer test was used when Chi-square test was not provided qualitative independent datain the analysis. The value of $p<0.05$ was considered statistically significant. The SPSS 22.0 softwarewas used forthe analysis.

RESULTS

Of 136 patient scheculed for liver transplantation, 127 were included (**Figure 1**). In the study, 127 patients in which 80 male (63%) and 47 female (37%) were scanned. As a result, statistically no significant differences were observed between the demographic characteristics and MELD scores of the patient groups (**Table 2**).

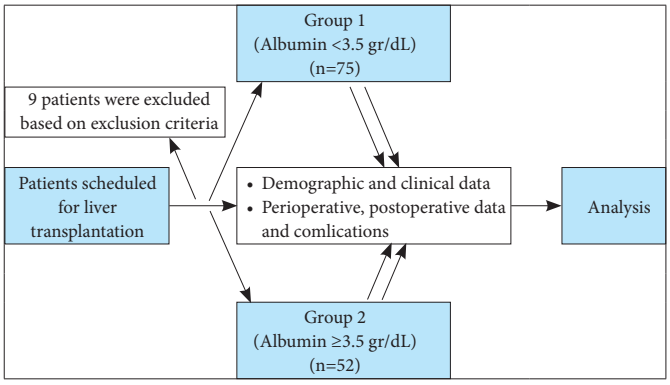

Figure 1. Flow diagram of study

The patients' postoperative mechanical ventilation times (weaning), intensive care unit stay, and time to discharge were compared, it was found that the durations were longer in the group with low albumin levels (**Table 2**).

Table 2. Demographic and clinical characteristics

	Group I		Group II		p
	Mean±SD /n-%	Median	Mean±SD /n-%	Median	
Demographic characteristics and MELD scores					
Age	54.2±12.0	57.0	53.3±13.0	57.0	0.693
Gender					
Male	49-65.3%		31–59.6%		0.512
Female	26-34.7%		21–40.4%		
MELD Skoru	21.1±4.6	20.0	20.7± 4.7	20.0	0.486
MV, ICU and Hospital stay					
MV duration (day)	2.3±1.4	2.0	1.2±0.4	1.0	0.000
ICU duration (day)	6.1±3.4	5.0	3.3±1.1	3.0	0.000
Length of hospital stay (day)	15.0±5.6	13.0	10.1±3.1	9.5	0.000
Use of blood and blood products					
ES	6.5±3.9	5.0	3.8±1.8	3.5	0.000
FFP	8.5±5.0	7.0	5.7±2.2	5.0	0.000
PS	2.1±1.3	2.0	2.1±1.3	2.0	0.936
Postoperative pulmonary complications					
Pleural effusion	50–66.7%		8–15.4%		0.000
Atelectasis	33–44.0%		2–3.8%		0.000
Pneumonia	23–30.7		2–3.8%		0.000
Creatinine levels					
Preoperative	0.78±0.26	0.7	0.73±0.23	0.7	0.279
Postoperative 1st day	1.03±0.49	0.9	0.74±0.28	0.7	0.000
Postoperative 2nd day	1.3±0.58	1.2	0.78±0.41	0.7	0.000
Acute Kidney Injury					
Stage I	29–38.7%		1–1.9%		0.000
Stage II	15–20%		2–3.8%		0.000
Stage III	5–6.7%		0–0.0%		0.000

MELD: Model for end-stage liver disease, MV: Mechanical ventilation, ICU: Intensive care unit ES: Erythroid suspension, FFP: Fresh frozen plasma, PS: Platelet suspension

In case of the blood and blood products used during operation in the patient groups, it was observed that the use of erythrocyte suspension and fresh frozen plasma was higher in the group with low albumin levels, and this difference was statistically significant too. Furthermore, there was no significant difference between the groups in the use of platelet suspension (**Table 2**).

The postoperative pulmonary complications were compared by examining the postoperative pulmonary radiographs and tracheal aspirate cultures of the patients, it was observed that the development of pleural effusion, atelectasis and pneumonia was significantly higher in patients with low serum albumin levels (**Table 2**).

The increase between the preoperative and postoperative creatinine levels of the patients werecompared. Consequently, a significant difference was observed between the groups (**Table 2**).

When the serum creatinine levels of the patients on the 1st and 2nd postoperative days were evaluated and acute kidney injuries were compared using the AKIN classification, it was observed that the development of stage I and stage II acute kidney injury was significantly higher in patients with low serum albumin levels (**Table 2**).

When the complications occuredin the early postoperative first 30 days of the patients were compared, it was observed that sepsis, acute kidney failure, unsuccessful weaning, bleeding-transfusion requirement, pneumonia, pulmonary edema, urinary tract infection, retransplantation and exitus development were significantly higher in patients with low serum albumin levels (**Table 3**).

Table 3. Early postoperative complications

	Grup I		Grup II		p
	n	%	n	%	
sepsis	21	28%	1	1.9%	0.000
Acute Renal Failure	30	40%	3	5.8%	0.000
Pneumonia	23	30.7%	2	3.8%	0.000
Bleeding / Transfusion	20	26.7%	2	3.8%	0.001
Failed weaning	11	14.7%	0	0%	0.004
Reintubation	4	5.3%	0	0%	0.144
Pulmoneredema	5	6.7%	0	0%	0.057
Urinary tract infection	15	20%	0	0%	0.001
Cardiac problems	8	10.7%	1	1.9%	0.059
Retransplantation	3	4.0%	0	0%	0.269
Eksitus	4	5.3%	0	0%	0.144

DISCUSSION

The frequency of hypoalbuminemia has been shown to be 30-40% in adult critically ill patients. Acute hypoalbuminemia as a result of albumin leakage into the interstitial space has been associated with high mortality, as the degree of capillary hyperpermeability is probably related to the degree of systemic inflammatory response (3). In patients with sepsis and other inflammatory conditions, the increased vascular permeability increases the transcapillary loss of albumin, participating into the development of hypoalbuminemia (4).

Albumin level is one of the main determinants of plasma oncotic pressure as well as reflecting nutritional status and being a negative acute phase reactant. In case of hypoalbuminemia, due to low oncotic pressure, it leaks into the interstitial space and causes hypovolemia. Therefore, hypovolemia developing during surgery in a patient with hypoalbuminemia requires more blood and blood product transfusions (5).

Many mechanisms cause hypoalbuminemia in patients who are followed up with the diagnosis of end-

stage liver failure and received liver transplantation. Decreased albumin synthesis, protein leakage to the interstitial area, blood loss in the perioperative period, relocation of the protein pool, peroperative and postoperative fluid resuscitation, and postoperative immunosuppressive agents are included in the etiology of hypoalbuminemia (6).

The most common complications in the postoperative period are pulmonary complications. The only laboratory test can predict the postoperative pulmonary complications that may develop in the preoperative period is the serum albumin value. The oxidation and degradation of albumin affect bioactive lipid mediators, which play an important role in antimicrobial defense and repair. Therefore, albumin level affects the development of postoperative infection, and the level of hypoalbuminemia is considered to be correlated with the severity of infection (7). Haskins et al. (8) reported that preoperative hypoalbuminemia increased the rate of pneumonia and reintubation in the postoperative period and caused prolonged mechanical ventilation in their study on patients with colon cancer. Yang et al. (9) also found in their study that hypoalbuminemia prolongs the duration of mechanical ventilation and it is one of the markers predicting the success of weaning in patients on mechanical ventilation support. It has been determined that, patients with hypoalbuminemia in the pediatric intensive care unit have longer mechanical ventilation periods and lower discharge rates from the intensive care unit. (10) It has been reported that, preoperative hypoalbuminemia increases the dependence on mechanical ventilator in patients undergoing coronary bypass surgery. (11), In another study, it was reported that, hypoalbuminemia increased all cardiac and pulmonary complications and the rate of emergency intubation. (12)

In this study, the rate of postoperative pulmonary complications (atelectasis, pleural effusion, pneumonia) was found to be higher in the patient group with low serum albumin levels. At the same time, weaning times were found to be longer in this group too. However, no significant difference was found in arterial blood gas samples taken to evaluate the postoperative pulmonary functions. It can be explained as the reason for this is close follow-up of mechanical ventilation settings and electrolyte values and the application of rapid replacement in order to keep the monitoring parameters within physiological limits.

Acute kidney injury is one of the major complications after liver transplantation. Its frequency is variable due to the variety of tests used in patient selection, classification, and evaluation of renal function. However, the common result in the studies shows that acute kidney injury and failure are common after liver transplantation which is found to be associated with increased cost, mortality, and morbidity. Wiederman et al. in the meta-analysis study (13), it was stated that serum albumin level could be used as an independent marker in the prediction of acute kidney injury, and it was also reported that the rate of acute kidney injury increased by 134% for each 1 g/dL decrease in serum albumin level. Preoperative hypoalbuminemia has been independently associated with acute kidney injury in non-cardiac surgeries. (14) Sang et al. it was shown in a study by AKI that postoperative hypoalbuminemia is associated with AKI. (15) In this study, it was found that 48-hour creatinine levels were higher in patients in the hypoalbuminemia group and the development of acute kidney injury was higher in these patients.

In patients with end-stage liver failure, hemostasis balance is impaired in favor of coagulopathy with physiopathological changes. When major surgery such as liver transplantation is added to the existing coagulopathy, massive blood transfusion is frequently appliedfor these patients. Although the use of blood has decreased continuously with the development of surgical and anesthesia techniques, better graft preservation, preoperative anemia treatment, better intraoperative monitoring of coagulation status, and pharmacological treatment of fibrinolysis, orthotopic liver transplantation often presents with an increased need for transfusion. Although it is difficult to predict the need for transfusion, studies have shown that the preoperative albumin level can be used to predict the amount of intraoperative transfusion. Kim et al. (5) reported that, preoperative hypoalbuminemia increased peroperative erythrocyte suspension transfusion in patients who underwent radical nephrectomy. Erdost et al. (16) in his study reported that, the use of intraoperative erythrocyte suspension and fresh frozen plasma increased in patients with preoperative hypoalbuminemia who developed acute kidney injury. Similarly, studies in which hypoalbuminemia and increased transfusion rate are reported in literature. (12,17) In this study, the rate of intraoperative erythrocyte suspension and fresh frozen plasma transfusion was found to be higher in the patient group with preoperative hypoalbuminemia.

Gibbs et al. reported that a decrease below 1 g/L in serum albumin level is associated with an increase in mortality rates from 1% to 29% and morbidity rates from 10% to 65% in non-cardiac surgeries. They also reported that hypoalbuminemia is a better indicator of some types of morbidity, especially sepsis and major infections. (1)

In the study in which the results were compared according to albumin levels of 30,676 patients who had colorectal cancer surgery, in the hypoalbuminemia group 30-day mortality was 3.7%, DVT 2.7%, pneumonia 4%, sepsis 4.1%, acute renal failure 0.9%, and these values were found to be significantly higher than the normal albumin level group (17). In another study, the 30-day mortality was found to be 16.3% in the hypoalbuminemia group, and significantly higher than the normal albumin level group (4.3%) (18). In the study, in which 204819 patient data from 16 major operations were included, postoperative 30-day mortality was 3.81%, pulmonary complications 8.78%, and sepsis 8.09% in the hypoalbuminemia group (19). In this study, sepsis was 28%, pneumonia was 30.7%, and 30-day mortality was 5.3%. It was thought that the high complication rates obtained were related to the long duration of the liver transplant operation, the high need for blood and blood products, and the longer length of stay in the intensive care unit and hospital.

CONCLUSION

As a result; Albumin is the most abundant protein in the plasma and undertakes many important functions. The measurement of this protein level, which is easily accessible and cost-effective, has become one of the routine laboratory tests evaluated preoperatively. In this study, the relationship between preoperative serum albumin level and postoperative mortality and morbidity were examined, and revealed that preoperative hypoalbuminemia causes many postoperative complications and is a strong indicator of early mortality. Based on these effects, it is concluded that albumin can be used as a guide and a valuable parameter in postoperative follow-up in the detection of perioperative and postoperative complications.

The retrospective nature and the inaccessibility of some patient files are the main limitations of the study.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was initiated with the approval of the Uludağ University Medical Faculty Clinical Researches Ethics Committee (Date: 2017, Decision No: 4/37).

Informed Consent: Because of retrospective design of the study, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

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Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version

REFERENCES

1. Gibbs J, Cull W, Henderson W, Daley J, Hur K, Khuri SF. Preoperative serum albumin level as a predictor of operative mortality and morbidity: results from the National VA Surgical Risk Study. Arch Surg 1999; 134: 36–42.
2. Carvalho JR, Verdelho Machado M. New Insights About Albumin and Liver Disease. Ann Hepatol. 2018;17: 547-60.
3. McCluskey A, Thomas AN, Bowles BJ, Kishen R. The prognostic value of serial measurements of serum albumin concentration in patients admitted to an intensive care unit. Anaesthesia 1996; 51: 724-7.
4. Nicholson JP, Wolmarans MR, Park GR. The role of albumin in critical illness. Br J Anaesth 2000; 85: 599–610
5. Kim K, Seo H, Chin JH, Son HJ, Hwang JH, Kim YK. Preoperative hypoalbuminemia and anemia as predictors of transfusion in radical nephrectomy for renal cell carcinoma: a retrospective study. BMC Anesthesiol 2015; 15: 103.
6. Rabie M, Negmi H, Hammad Y, Oufi H, Khalaf H. Living donor hepatectomy: comparative study between two different anesthetic techniques MEJAnesth2006; 18: 4
7. Wiedermann CJ. Hypoalbuminemia as Surrogate and Culprit of Infections. Int J Mol Sci. 2021; 22: 4496
8. Haskins I, Baginsky M, Amdur R, Agarwal A. Preoperative hypoalbuminemia is associated with worse outcome in colon cancer patiets. ClinNutr 2016; 12: 1-6.
9. Yang P-H, Hung J-Y, Yang C-J. Successful weaning predictors in a respiratory care center in Taiwan. Kaohsiung J Med Sci 2008; 24: 85-91.
10. Leite HP, Rodrigues da Silva AV, de Oliveira Iglesias SB, Koch Nogueira PC. Serum albumin is an independent predictor of clinical outcomes in critically ill children.Pediatr Crit Care Med 2016; 17: e50-7.
11. Ji Q, Duan Q, Wang X, et al. Risk factors for ventilator dependency following coronary artery bypass grafting. Int J Med Sci 2012; 9: 306-10.
12. Lee D, Lee R, Cross MT, Tran A, Kappa J, Moghtaderi S. Low serum albumin levels are associated with increased 30-day cardiopulmonary complications, reoperation, and readmission rates following total shoulder arthroplasty. Iowa Orthop J 2019; 39: 27-34
13. Wiederman C, Wiederman W, Joannidis M. Hypoalbuminemia and acute kidney injury: a meta-analysis of observational clinical studies. Intensive care med 2010; 36: 1657-65.
14. Li N, Qiao H, Guo JF, et al. Preoperative hypoalbuminemia was associated with acute kidney injury in high-risk patients following non-cardiac surgery: a retrospective cohort study. BMC Anesthesiol 2019; 19: 171.
15. Sang BH, Bang JY, Song JG, Hwang GS. Hypoalbuminemia within two postoperative days is an independent risk factor for acute kidney injury following living donor liver transplantation: a propensity score analysis of 998 consecutive patients. Crit Care Med 2015; 43: 2552–61.
16. Erdost H, Özkardeşler S, Akan M, et al. Comprasion of the RIFLE, AKIN and KDIGO. Diagnosticclassifications for acute renal injury in patients undergoing liver transplantation. Transplant Proc 2015; 47: 1482-7.

17. Hu WH, Eisenstein S, Parry L, Ramamoorthy S. Preoperative malnutrition with mild hypoalbuminemia associated with postoperative mortality and morbidity of colorectal cancer: a propensity score matching study. Nutr J 2019; 18: 33.
18. Jellinge ME, Henriksen DP, Hallas P, Brabrand M. Hypoalbuminemia is a strong predictor of 30-day all-cause mortality in acutely admitted medical patients: A prospective, observational, cohort study. PLoS One 2014; 9: e105983.
19. Meyer CP, Rios-Diaz AJ, Dalela D, et al. The association of hypoalbuminemia with early perioperative outcomes - A comprehensive assessment across 16 major procedures. Am J Surg 2017; 214: 871-83.

Case Report

ANATOLIAN CURRENT MEDICAL

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Anatolian Curr Med J 2022; 4(3); 323-325

Sigmoid colon duplication seen as a rare cause of ileus in adult: case report

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ABSTRACT

Intestinal duplications are congenital anomalies usually seen in infancy. Since it is congenital, they often become symptomatic in childhood. Usually diagnosed by adulthood. Those seen in childhood are mostly seen in the part from the oral cavity to the middle of the transverse colon, which is defined as the foregut and midgut embryologically. In our case report, we presented a female patient who was diagnosed both in adulthood and presented with ileus caused by duplication in the sigmoid colon. A 24-year-old female patient was admitted with the complaints of intermittent constipation and inability to pass gas and stool for 3 days. It was thought that the pathology observed in the sigmoid colon in the abdominal CT with contrast was due to torsion. Urgent surgery was decided for the patient. During the operation, it was observed that there was duplication in the sigmoid colon and torsion developed due to this. The patient underwent anterior resection and end-to-end anastomosis. She was discharged with surgical recovery in the postoperative period. Intestinal duplications are congenital anomalies that are usually seen in infancy. However, although rarely, it can be detected in adulthood, as in the patient we presented. Duplication anomalies should be kept in mind in the differential diagnosis of patients with chronic constipation and abdominal pain.

Keywords: Sigmoid colon, duplication, ileus

INTRODUCTION

Intestinal duplications are congenital anomalies that are seen with a high rate under 2 years of age. It is seen in approximately 0.005-0.025% of births (1). It is usually observed in the intestinal tract up to the middle of the transverse colon and rarely seen in the sigmoid colon (2). Duplications may be cystic or tubular in nature. The patient's complaints vary according to the location of the duplication (3). It may present with acute or chronic symptoms. Preoperative diagnosis is difficult. A pathognomic finding cannot be found with imaging methods. Definitive treatment is surgery. Definitive diagnosis can be made by pathological examination of the resected intestinal segment. In our case report, we planned to present a 24-year-old female patient who was seen in adult age and presented with ileus, and to discuss the diagnosis and treatment of intestinal duplication.

CASE REPORT

A 24-year-old female patient presented to the emergency department with complaints of abdominal pain, nausea, vomiting and inability to pass gas and stool. Except that she had occasional constipation and abdominal pain in her history, there was no feature in her history and family history. On physical examination, there was abdominal distension and tenderness in the lower quadrants. There were no pathological findings except leukocytosis and CRP elevation in laboratory results. As a result of physical examination and tests done for the patient, sigmoid colon volvulus was suspected on computed tomography (**Figure 1**). Emergency surgery was decided for the patient. Peroperatively, it was observed that the possible duplication of the sigmoid colon, which ended with a blunt tip, which was approximately 40 cm long and reached a diameter of 10 cm at its widest point, caused obstruction and the proximal intestinal loops were dilated. No perforation observed. Resection and end-to-end anastomosis were performed to this structure and the sigmoid colon to which it is associated (**Figure 2**). The patient, who had no problems in the postoperative follow-up, was discharged with surgical recovery. Pathological examination of the surgical specimen was consistent with sigmoid colon duplication.

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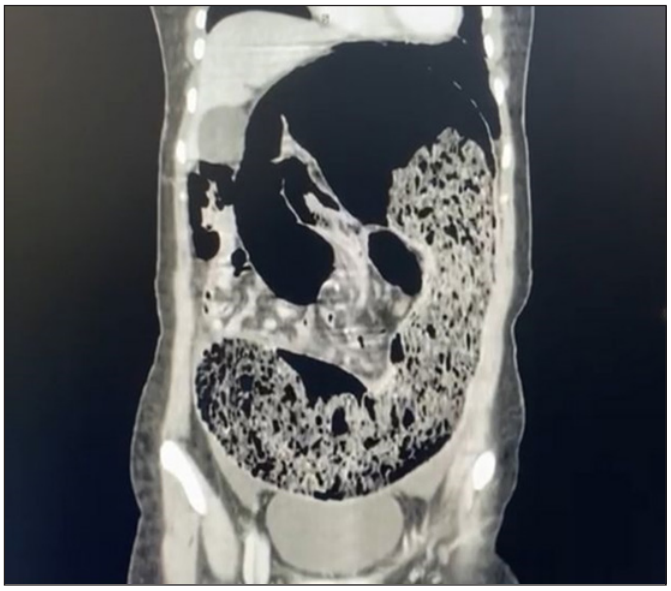

Figure 1. Volvulus-like image on tomography

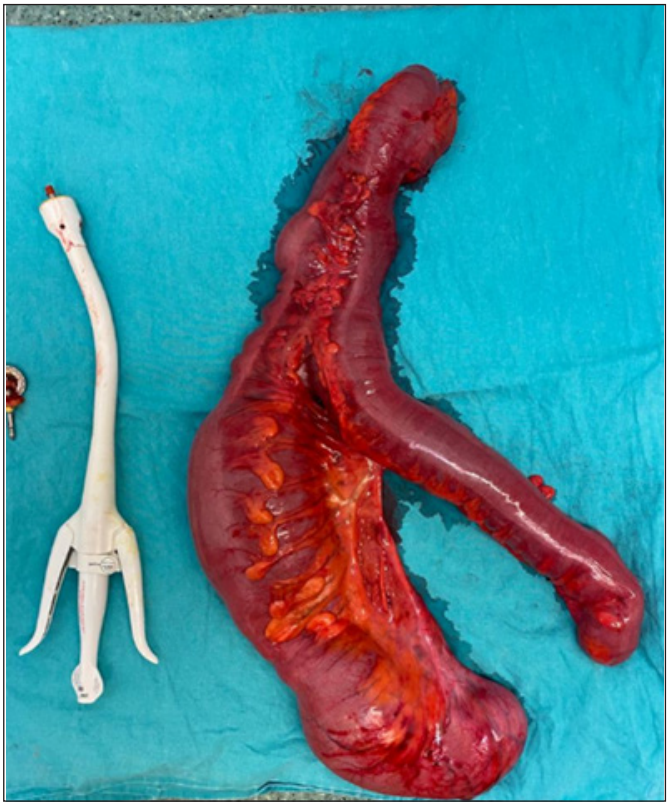

Figure 2. Sigmoid colon and duplication resection material

DISCUSSION

Gastrointestinal system duplications are very rare congenital anomalies that can occur in any part of the digestive tract. It is seen in approximately 0.005-0.025% of births. Diagnosis rate has increased with the use of intrauterine ultrasonography. Symptoms are observed in 67-80% of patients, mostly before the age of 2 years. Although it is seen in the abdomen at a rate of 80%, it can also occur in the thorax. Duplications seen in the digestive tract are most frequently observed in the ileum (30-35%) and the least in the colon (7-20%) (4 -6). In the colon, it is most commonly observed in the transverse colon (3). Duplications may be cystic (80%) or tubular (20%) structurally (2,7). The low incidence of tubular colonic duplications explains that most patients remain asymptomatic until adulthood, as was the case in our patient. The patient we presented had a 24-year-old sigmoid colon duplication presenting with ileus.

Classification of gastrointestinal duplications depends on their morphology and region of origin. In 1969, McPherson et al. (8) classified for colonic duplications. Accordingly, type 1 is simple cystic, type 2 is diverticular, type 3 is tubular colonic duplication. This classification can also be used to evaluate the duplication structurally. Tubular sigmoid colon duplication was also detected in the patient we presented.

Symptoms of intestinal duplications may vary depending on the region and type of duplication. Abdominal pain, constipation, swelling in the abdomen, rectal bleeding, volvulus are the symptoms and signs that can be seen. Although less common, Kang M et al. (9). It has also been reported that malignancy can also develop on the basis of duplication.

There is no definite finding that can diagnose intestinal system duplications with imaging methods. Even in duplications in the colonic tubular structure, duplication may be overlooked during colonoscopy due to the narrow junction with the colon or the fact that it is covered with stool. Contrast-enhanced abdominal tomography and barium enema can provide more information than other methods. However, there is still no definitive preoperative imaging method. In patients with chronic symptoms, surgery is important in terms of elimination of symptoms and diagnosis. Since some of the patients present with the clinic of acute abdomen, the diagnosis can be made during the operation and with the pathological examination in the postoperative period. In the case we presented , the patient who had ileus in the preoperative period was thought to have sigmoid colon volvulus as a result of tests . The tomography finding was that there may be sigmoid colon duplication, cyst, mass and volvulus. In the exploration, sigmoid colon torsion was observed due to duplication in the sigmoid colon. Accordingly, the proximal intestinal loops were dilated. The patient underwent anterior resection and end-to-end anastomosis, including duplication. Pathological evaluation was consistent with sigmoid colon duplication. No malignancy was detected.

CONCLUSION

Most of the duplications in the gastrointestinal tract are seen and treated under the age of 2 years. As in the rare case we presented, it may remain asymptomatic until adulthood and present with acute or chronic symptoms. Intestinal duplications should be kept in mind in adults with symptoms such as abdominal pain and constipation or in the differential diagnosis of bowel obstructions.

ETHICAL DECLARATIONS

Informed Consent: Written informed consent was obtained from all participants who participated in this study.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

Financial Disclosure: The authors declared that this study has received no financial support.

Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper and that they have approved the final version

REFERENCES

1. Polydorides AD. Colon: colonic duplications, cysts, and congenital diverticula. In: Greenson JK, editor. Diagnostic pathology: gastrointestinal. 3rd ed. London: Elsevier Health Sciences; 2019. p. 364-5. Section 5.
2. Mourra N, Chafai N, Bessoud B, Reveri V, Werbrouck A, Tiret E. Colorectal duplication in adults: report of seven cases and review of the literature. J Clin Pathol 2010; 63: 1080-3.
3. Wu X, Xu X, Zheng C, Li B. Tubular colonic duplication in an adult: case report and brief literature review. J Int Med Res 2018; 46: 2970-5.
4. Heiss K. Intestinal duplications. In: KT Oldham, PM Colombani and RP Foglia (eds). Surgery of infants and children: Scientific principles and Practice. Philadelphia: Lippincott–Raven, 1997, pp.1265-76.
5. Puligandla PS, Nguyen LT, St-Vil D, et al. Gastrointestinal duplications. J Pediatr Surg 2003; 38: 740-4.
6. Holcomb GW 3rd, Gheissari A, O'Neill JA Jr, Shorter NA, Bishop HC. Surgical management of alimentary tract duplications. Ann Surg 1989; 209: 167-74.
7. Kekez T, Augustin G, Hrstic I, et al. Colonic duplication in an adult who presented with chronic constipation attributed to hypothyroidism. World J Gastroenterol 2008; 14: 644-6
8. McPherson AG, Trapnell JE and Airth GR. Duplication of the colon. Br J Surg 1969; 56: 138-42.
9. Kang M, An J, Chung DH, Cho HY. Adenocarcinoma arising in a colonic duplication cyst: a case report and review of the literature. Korean J Pathol 2014; 48: 62-5.

Original Article

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ERRATUM

The validity and agreement of PI-RADS v2 in the diagnosis of prostate cancer

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ERRATUM

In the article with "**Tezcan Ş, Bekar Ü, Gürbüz Onbaşıoğlu M, Ergin G. The validity and agreement of PI-RADS v2 in the diagnosis of prostate cancer. Anatolian Curr Med J 2021; 3(4); 303-309**" citation information which was published (Date: 24.09.2021) in the fourth issue of third volume of Anatolian Current Medical Journal, authors noticed a misreport in the number of the included and excluded patients in the "Material and Method" section and **Figure 1**. Authors apologize to the readers for the mistake. In this paper, additional explanations and corrections are reported to remedy the mistake.

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ERRATUM

1. The number of included and excluded patients in the study was inadvertently misspelled in the section of "Study Population" in the "Material and Method" on page 304. It is understood that some mistakes were made inadvertently in the writing of the sentences which were "146 patients with clinically suspected PC based on blood PSA or clinical examination with no prior biopsy or with prior negative biopsy who were underwent mpMRI between January 2017 and January 2020 were enrolled in this study. Of these patients, 41 patients who had not histopathologic evaluation in our hospital were excluded from this study." in this section. In those sentences, the total number of patients must be written as 246 patients instead of 146 patients and the number of excluded patients must be written as 141 patients instead of 41 patients. As a result, the sentences should have been as follows: "246 patients with clinically suspected PC based on blood PSA or clinical examination with no prior biopsy or with prior negative biopsy who were underwent mpMRI between January 2017 and January 2020 were enrolled in this study. Of these patients, 141 patients who had not histopathologic evaluation in our hospital were excluded from this study.". Consequently, those corrections do not affect the number of patients ultimately included, statistical analysis, results and conclusion of this study.
2. The number of included and excluded patients was also inadvertently misspelled in the flowchart showing the included and excluded patients in **Figure 1** on page 305. This flowchart was also corrected and the accurate form of the flowchart was shown in **Figure 1** below the text.

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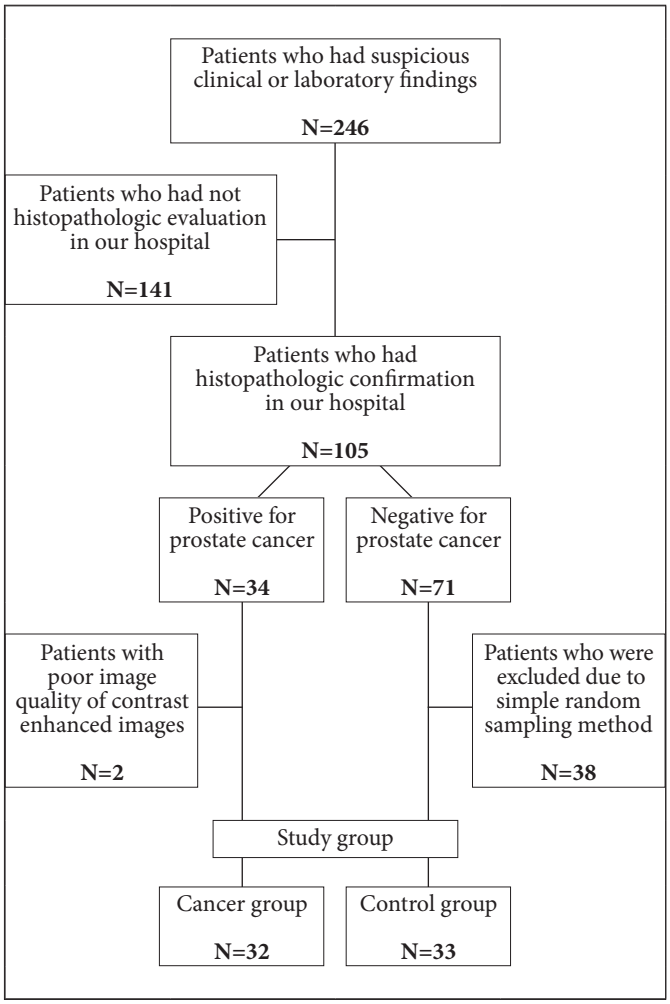

Figure 1. Flowchart of inclusion and exclusion criteria of study sample

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