ARAŞTIRMA MAKALESİ/ORIGINAL RESEARCH

DOI: 10.5505/ktd.2022.93546
Kocaeli Med J 2022;11(3)254-302

Association of Changes in Inflammatory Markers with Mortality in COVID-19 Infection Treated with Tocilizumab

Tosilizumab ile Tedavi Edilen COVID-19 Enfeksiyonunda İnflamatuar Belirteçlerdeki Değişikliklerin Mortalite ile İlişkisi

Mustafa Tanrıverdi[1], Nevhiz Gündoğdu[2]

1SANKO Üniversitesi Tıp Fakültesi, Enfeksiyon Hastalıkları ve Klinik Mikrobiyoloji Anabilim Dalı, Gaziantep,Türkiye
2SANKO Üniversitesi Tıp Fakültesi, Göğüs Hastalıkları Anabilim Dalı, Gaziantep, Türkiye

ÖZ

GİRİŞ ve AMAÇ**:**Tosilizumabın ciddi COVID-19 enfeksiyonunda mortaliteyi azalttığı bilinmektedir. Biz, tosilizumab ile tedavi edilen COVID-19 enfeksiyonu olan hastalarda mortalite ile ilişkili faktörleri ve klinik ve inflamatuar bulgulardaki değişiklikleri incelemeyi amaçladık.

YÖNTEM ve GEREÇLER: Tosilizumab ile tedavi edilen COVID-19 enfeksiyonu olan erişkin hastalar retrospektif olarak analiz edildi. Glukokortikoide cevap vermeyen makrofaj aktivasyon sendromu saptanan veya klinik seyri hızlı ilerleyen COVID-19 enfeksiyonunda tosilizumab endikedir. Bazal (tosilizumab öncesi) ve tosilizumab sonrası 1., 3. ve 5. günlerdeki klinik ve laboaratuar parametrelerindeki değişikliği karşılaştırdık. Tosilizumab tedavisinin 30.gününde mortalite gelişimine göre hastaları grupladık: yaşayanlar ve eksituslar.

BULGULAR: Hastaların ortalama yaşı 61.54 (±10.27) ve toplamın (n=76) %28.9'u (n=22) kadındı. 14 hasta (%18.42) öldü. Lenfosit sayısı (≤170), SaO2(≤2), D-dimer (>0.18), lökosit (>2400), prokalsitonin (≤-0.11), LDH (>90) ve ALT(≤11) seviyelerinde bazal ve tedavinin 5.günü arasındaki fark mortaliteyi predikte ettirdi. Kaplan-Meier analizinde, ateş ve anorekside azalma, tedavinin 5.günü ile bazal SaO2, nötrofil, lenfosit ve LDH seviyeleri arasındaki fark mortaliteyi predikte ettirdi. Cox regresyon analizinde ateşin devamlılığı, D-dimer lökosit ve nötrofil düzeylerindeki artışın mortalitenin önemli prediktörleri olduğu saptandı.

TARTIŞMA ve SONUÇ: Mortalite oranı rölatif olarak düşüktü. Ateşin devamlılığı, lökosit, nötrofil ve D-dimer düzeylerinde artış mortaliteyi predikte ettirdi. Tosilizumab tedavisi altındaki COVID-19 enfeksiyonu olan hastalarda inflamatuar belirteçlerin ölçülmesini öneriyoruz

Anahtar Kelimeler: COVID 19, tosilizumab, SARS CoV-2, inflamasyon, mortalite.

ABSTRACT

INTRODUCTION: Tocilizumab is known to reduce mortality in severe COVID-19 infection. We aimed to investigate the changes in clinical and inflammatory findings, and the factors associated with mortality in patients with COVID-19 infection treated with tocilizumab.

METHODS: Adult patients with COVID-19 infection treated with tocilizumab were analyzed retrospectively. Tocilizumab was indicated in COVID-19 infection and macrophage activation syndrome, which was unresponsive to glucocorticoids, or where the clinical course had progressed rapidly. We compared changes in the clinical and laboratory parameters between baseline (pre-tocilizumab), post-tocilizumab 1st day, 3rd day, and 5th day periods. We grouped the patients according to mortality at the 30th day of treatment with tocilizumab alive vs. exitus.

RESULTS: The mean age of patients was 61.54 (±10.27), 28.9% (n=22) of the total (n=76) were female. 14 patients (18.42%) had died. The change in lymphocyte count (≤170), and SaO2(≤2), D-dimer (>0.18), leukocyte (>2400), procalcitonin (≤-0.11), LDH (>90), and ALT(≤11) levels between the 5th day after treatment and the baseline predicted mortality. Kaplan-Meier analysis showed that the decrease in fever and anorexia, the change in SaO2, neutrophil, lymphocyte, and LDH levels between the 5th day after treatment and the baseline were predictors for mortality. Cox regression analysis revealed that persistence of fever, and an increase in D-dimer, leukocyte, and neutrophil levels were significant predictors for mortality.

DISCUSSION AND CONCLUSION: The mortality rate was relatively low. Persistence of fever, increase in leukocyte, neutrophil or D-dimer levels predicted mortality. We recommend measurement of inflammatory markers in the patients COVID-19 infection under treatment of tocilizumab.

Keywords: COVID 19, tocilizumab, SARS CoV-2, inflammation, mortality.

Kabul Tarihi: 29.12.2022

Correspondence: Aynur Koyuncu Hasan Kalyoncu Üniversitesi Sağlık Bilimleri Fakültesi, Gaziantep, Türkiye

E-mail: aynrkoyuncu@yahoo.com.tr

INTRODUCTION

COVID-19 was declared a pandemic by the World Health Organization (WHO) on March 11th, 2020, and has affected about 6.5 million people worldwide up to September 2022 (1). Several studies have shown that SARS-CoV-2 infection resulted in a high mortality rate in the general population (1, 2). The Ministry of Health of Türkiye declared that more than 15 million people had been affected by the SARS-CoV-2 infection, and that more than 100,000 people had died up to September 2022 (3).

Although most of the patients infected with SARS-CoV-2 had complained of minor symptoms, hospitalization, oxygen support and intensive care unit admission were necessary in some cases (4). The higher mortality rates in COVID-19 infection were associated with obesity, diabetes mellitus, cancer, or aging (5-8). After mass vaccination programs had begun, the rate of severe infection, and the mortality rates of COVID-19 infection declined (1, 5, 9-13).

Cytokine storm and macrophage activation were shown to be important factors in the pathogenesis of the SARS-CoV-2 infection (14). In previous reports, mortality in COVID-19 infection was shown to be associated with various factors such as oxygen saturation at admission, CRP, interleukin-6, procalcitonin, D-dimer levels, or leukocyte count (15-21). Because IL-6 has been considered one of the most important cytokines involved in cytokine release syndrome, anti-IL-6 agents have been proposed as a treatment for COVID-19 infection. Tocilizumab, an anti-IL-6 agent, was shown to affect the prognosis and decrease mortality rates in severe COVID-19 infection (4,22-24).

We aimed to investigate the changes in clinical and laboratory parameters with tocilizumab treatment, and the factors associated with mortality in patients with COVID-19 infection treated with tocilizumab.

MATERIALS AND METHODS

Study Design

This retrospective study was conducted in SANKO University Medical Faculty hospital, and approved by the local SANKO University Clinical Research Ethics Committee with approval number of 04 (session no: 2021/06). The study was conducted in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments. Written informed consent was obtained from all of the participants.

The adult patients diagnosed and followed-up with COVID-19 infection and treated with tocilizumab in SANKO University Medical Faculty hospital between July 2020 and December 2020 were analyzed retrospectively. COVID-19 diagnosis was made by PCR test analyzed from nasal swab samples taken from patients in whom there was a suspicion of COVID-19 infection. All patients underwent clinical and radiological evaluation. Those for whom data were missing were not included in the study.

Data Collection

Demographic parameters (age, sex, and body mass index [BMI], blood group), clinical parameters (symptoms, chronic illnesses [hypertension, type 2 diabetes mellitus, asthma, chronic obstructive pulmonary disease, and coronary artery disease], oxygen saturation (SaO2, %), duration of hospitalization [days]), and laboratory findings (C-reactive protein [CRP], ferritin, D-dimer, procalcitonin, fibrinogen, alanine transaminase [ALT], aspartate aminotransferase [AST], lactate dehydrogenase [LDH] and troponin levels, and lymphocyte, neutrophil [PMNL] and leukocyte [WBC] counts) were recorded by using electronic and written patient files.

Complete blood count was studied with an automated analyzer.

Patient Groups

The patients were grouped based on clinical symptoms and pulmonary imaging findings on thorax computed tomography (CT) on admission. We grouped them according to the severity of the infection as Group A - outpatient (mild symptoms of COVID-19 infection, but no CT findings); Group B - mild/moderate illness (fever and respiratory tract symptoms of COVID-19 infection together with pneumonia on CT); Group C - severe (respiratory rate >30/minute, or oxygen saturation at room air of <93%, or PaO2/FiO2 of <300, or an increase in CT findings of more than 50% in 1-2 days) or critical illness (shock, or respiratory insufficiency requiring mechanical ventilation, or organ failure necessitating intensive care).

The patients, who were grouped in Group A at diagnosis, but who later became increasingly ill, were re-classified as either Group B or C.

Tocilizumab treatment

Tocilizumab was indicated in those patients with macrophage activation syndrome developed during the COVID-19 infection, who were unresponsive to glucocorticoid treatment or whose clinical course was rapidly progressive (25).400 mg of tocilizumab was administered intravenously in a single dose, which was followed by a second dose of 200-400 mg in the 24th hour after the first dose in some patients (25). Tocilizumab was not given to those patients with contraindications such as pregnancy, neutropenia (<500/mm3), active tuberculosis, hepatitis B or C infections, hypersensitivity, or elevated liver enzymes (≥5-fold).

We compared changes and differences in the clinical and laboratory parameters between baseline (pre-tocilizumab, before tocilizumab treatment), post-tocilizumab treatment 1st day, post-tocilizumab treatment 3rd day, and post-tocilizumab treatment 5th day periods.

At first all patients were treated with glucocorticoids. Glucocorticoids were continued in those patients, who were treated with tocilizumab.

We analyzed the mortality in the 1st 30 days of treatment with tocilizumab, and we grouped the patients according to mortality at the 30th day of treatmnet: alive vs. exitus. We compared demographic, clinical and laboratory parameters between the groups.

Statistical Analysis

Data obtained in the study were analysed statistically using SPSS 25.0 (IBM Corporation, Armonk, New York, United States) and PAST 3(Hammer, Ø., Harper, D.A.T., Ryan, P.D. 2001. Paleontological statistics). The conformity of the univarible data to normal distribution was evaluated using the Shapiro-Wilk francia test, and homogenity of variance was evaluated with the Levene test. The conformity of the multivarible data to normal distribution was evaluated using the Mardia (Dornik and Hansen omnibüs) test, and homogenity of variance was evaluated with the Box-M test. When comparing two independent groups of quantitative data according to each other, we used the nonparametric Mann-Whitney U test with Monte Carlo results. In the comparison of the parameters of fever, cough, dyspnea and anorexia between pre-tocilizumab, post-tocilizumab 1st, 3rd and 5th day periods, the Cochran Q Test with Monte Carlo simulation results, and Dunn's test for posthoc tests, was used. In the comparison of levels and counts with respect to oxygen saturation, CRP, ferritin, D-dimer, white blood cell, neutrophil, lymphocyte, procalcitonin, fibrinogen, LDH, troponin, AST and ALT between pre-tocilizumab, and post-tocilizumab 1st, 3rd, and 5th day periods, the Friedman Test with Monte Carlo simulation results was used, and a Stepwise step-down comparisons test was used for Post-hoc tests. When comparing categorical variables with each other, the Pearson Chi-square and Fisher exact tests with Monte Carlo simulation technique were used. To detect the relationship between the real classification of the procedure's success and the classification made by the cut-off values, sensitivity and specificity ratios, and positive and negative predictive values were expressed by ROC (Receiver Operating Curve) curve analysis. Kaplan-Meier (product limit method) Log Rank (Mantel-Cox) analysis was used to evaluate the effect of the factors on survival and mortality. To measure the effects of prognostic variables on mortality, Cox regression analysis with the Backward Stepwise (Wald) method was used. Quantitative variables were stated as mean (standard deviation), and median (minimum-maximum) values, and categorical variables as number (n) and percentage (%) in the tables. Variables were evaluated at a 95% confidence level, and a value of $p<0.05$ was accepted as statistically significant.

RESULTS

The mean age of patients was 61.54 (±10.27), 28.9% (n=22) of the total (n=76) were female. 14 patients (18.42%) had died. Age, sex, blood group, severity, and comorbidities were not different in either the exitus or alive groups (Table 1). Fever, cough, dyspnea, anorexia and fatigue were present in all patients at the baseline.

The ratio of those patients with fever had begun to decrease during the 3rd day of tocilizumab treatment, and continued to decrease during the 5th day in the whole group. The ratio of those patients with anorexia decreased 5 days after tocilizumab treatment, however, cough or dyspnea persisted after treatment. SaO2 and lymphocyte count increased, and CRP, procalcitonin and fibrinogen decreased after treatment in the whole group. D-dimer, PMNL count and LDH levels did not significantly change with treatment. Ferritin levels first increased during the 1st day of treatment then

decreased to pretreatment levels. AST levels did not consistently increase, but ALT levels increased after treatment (Tables 2, 3 and 4).

Decrease in fever and anorexia was more frequent in 5th day after tocilizumab treatment in the alive group than in the exitus group. Baseline SaO2 levels were lower, and D-dimer and LDH levels higher in the exitus group than those in the alive group. SaO2 levels were higher after treatment in the alive group, and D-dimer levels increased in the exitus group. The SaO2 change was higher in the alive group than that in the exitus group ($p<0.001$). D-dimer change was lower in the alive group than that in the exitus group ($p=0.016$). Change in CRP, WBC, ferritin, PMNL, troponin or fibrinogen levels were similar in both the alive and exitus groups. Procalcitonin levels at the baseline and in the 1st, 3rd, and 5th days after tocilizumab treatment were lower in the alive group than those in the exitus group. Changes in ALT and AST levels in the 5th day after tocilizumab treatment were higher in the alive group than those in the exitus group (Tables 5, 6, 7, and 8).

Changes in SaO2 (≤2), D-dimer (>0.18), PMNL (>2400), procalcitonin (≤-0.11), LDH (>90), and ALT (≤11) levels as well as lymphocyte count (≤170 between the 5th day after treatment and the baseline were associated with mortality (Table 9).

Kaplan-Meier analysis showed that persistence of fever and anorexia, changes in SaO2, PMNL, lymphocyte, and LDH levels between the 5th day after treatment and the baseline were associated with mortality (Tables 10 and 11).

Cox regression analysis revealed that persistence of fever, and an increase in D-dimer, WBC, and PMNL levels were significant predictors for mortality (Table 12).

Table 1. Baseline Characteristics of the Patients

	Total (n=76)	Exitus (n=14)	Alive (n=62)	p value
	n(%) or median (min.-max.)	n(%) or median (min.-max.)	n(%) or median (min.-max.)	
Sex (female)	22(28.9)	5 (35.7)	17 (27.4)	0.531 [f]
Age	63(38-79)	67 (46 / 73)	61 (38 / 79)	0.201 [u]
BMI	28.5(23-60)	28 (23 / 38)	29 (23 / 60)	0.904 [u]
Blood group				0.887 [ff]
Zero	19(25.0)	4 (28.6)	15 (24.2)	
A	32(42.1)	6 (42.9)	26 (41.9)	
B	19(25.0)	4 (28.6)	15 (24.2)	
AB	6(7.9)	0 (0)	6 (9.7)	
Severity (severe)	67(88.2)	14 (100)	53 (85.5)	0.197 [f]
Co-Existent illnesses	56(73.7)	13 (92.9)	43 (69.4)	0.097 [f]
Hypertension	34(44.7)	9 (64.3)	25 (40.3)	0.139 [c]
T2D	36(47.4)	8 (57.1)	28 (45.2)	0.556 [c]
Asthma	5(6.6)	1 (7.1)	4 (6.5)	0.999 [f]
COPD	4(5.3)	1 (7.1)	3 (4.8)	0.565 [f]
CAD	12(15.8)	3 (21.4)	9 (14.5)	0.685 [f]
Sputum	4(5.3)	1 (7.1)	3 (4.8)	0.565 [f]
Duration of hospitalization	14(6-43)	17 (8/43)	14 (6/39)	0.098 [u]

[f] Fisher Exact Test (Monte Carlo), [ff] Fisher Freeman Halton test (Monte Carlo),

[c] Pearson Chi Square Test (Monte Carlo), [u] Mann Whitney U Test (Monte Carlo),

min: Minimum, max: Maximum T2D: type 2 diabetes mellitus COPD: chronic obstructive pulmonary disease CAD: coronary artery disease

Table 2. Changes in Clinical Findings with Tocilizumab Treatment

	Fever	Cough	Dyspnea	Anorexia	SaO2
	n (%)	n (%)	n (%)	n (%)	Median (min / max)
PRET.	76 (100)	76 (100)	75 (98.7)	76 (100)	88 (85 / 92)
POSTT. 1. day	76 (100)	76 (100)	76 (100)	76 (100)	89 (86 / 92)
POSTT. 3. day	42 (55.3)	76 (100)	75 (98.7)	63 (82.9)	91 (86 / 93)
POSTT. 5. day	6 (7.9)	76 (100)	72 (94.7)	26 (34.2)	93 (86 / 95)
p (intragroup)	**<0.001** [c]	-	0.140 [c]	**<0.001** [c]	**<0.001** [fr]
PRET. vs POSTT. 1. Day	-	ns.	ns.	-	**<0.001**
PRET. vs POSTT. 3. Day	**<0.001**	ns.	ns.	0.076	**<0.001**
PRET. vs POSTT. 5. Day	**<0.001**	ns.	ns.	**<0.001**	**<0.001**
POSTT. 1. Day vs POSTT. 3. Day	**<0.001**	ns.	ns.	0.076	**<0.001**
POSTT. 1. Day vs POSTT. 5. Day	**<0.001**	ns.	ns.	**<0.001**	**<0.001**
POSTT. 3. Day vs POSTT. 5. Day	**<0.001**	ns.	ns.	**<0.001**	**0.001**

[fr] Friedman Test(Monte Carlo); Posthoc Test: Stepwise step-down comparisons), [c] Cochran's Q test(Monte Carlo); Post Hoc Test : Dunn's Test
min: Minimum, max: Maximum, ns.: Not significant, PRET.:Before tocilizumab, POSTT.:After tocilizumab SaO2: oxygen saturation

Table 3. Changes in Laboratory Findings with Tocilizumab Treatment

	CRP	Ferritin	D-dimer	WBC	PMNL	Lymphocyte
	Median (min / max)	Median (min / max)	Median (min / max)	Median (min / max)	Median (min / max)	Median (min / max)
PRET.	124 (40 / 350)	761.5 (83 / 2000)	0.81 (0.27 / 5.54)	8090 (1950 / 20240)	7235 (1000 / 18190)	640 (180 / 1600)
POSTT. 1. Day	80 (15 / 315)	841 (93 / 2000)	0.97 (0.27 / 20)	8855 (2620 /	7580 (1900 /	813 (110 / 2160)
POSTT. 3. Day	20 (1 / 126)	777.5 (154 / 2000)	1 (0.12 / 20)	9065 (2860 /	7315 (1720 /	975 (130 / 3000)
POSTT. 5. Day	6 (1 / 68)	717 (65 / 2000)	0.98 (0.1 / 3.87)	9095 (1060 /	7575 (2300 /	1280 (180 / 3940)
P (intragroup)	**<0.001**	**<0.001**	0.073	**<0.001**	0.534	**<0.001**
PRET. vs POSTT. 1. Day	**<0.001**	**0.023**	ns.	0.999	ns.	**0.121**
PRET. vs POSTT. 3. Day	**<0.001**	0.999	ns.	**0.072**	ns.	**<0.001**
PRET. vs POSTT. 5. Day	**<0.001**	0.999	ns.	**0.001**	ns.	**<0.001**
POSTT. 1. Day vs POSTT. 3. Day	**<0.001**	**0.212**	ns.	0.614	ns.	**0.015**
POSTT. 1. Day vs POSTT. 5. Day	**<0.001**	**0.001**	ns.	**0.015**	ns.	**<0.001**
POSTT. 3. Day vs POSTT. 5.	**<0.001**	**0.411**	ns.	0.999	ns.	**0.003**

Friedman Test(Monte Carlo); Posthoc Test: Stepwise step-down comparisons),
min: Minimum, max: Maximum, ns.: Not significant, PRET.:Before tocilizumab, POSTT.:After tocilizumab CRP: C-reactive protein WBC: white blood cell PMNL: polymorphonuclear leukocyte

Table 4. Changes in Laboratory Findings with Tocilizumab Treatment

	Procalcitonin	Fibrinogen	LDH	Troponin	AST	ALT
	Median (min / max)	Median (min / max)	Median (min / max)	Median (min / max)	Median (min / max)	Median (min / max)
PRET.	0.07 (0.02 / 1.07)	713.5 (383 / 1200)	402 (132 / 1077)	3 (1 / 120)	34 (8 / 152)	36.5 (8 / 308)
POSTT. 1. Day	0.05 (0.01 / 0.75)	650.5 (309 /	387 (132 / 1053)	4 (1 / 82)	36 (8 / 177)	46 (8 / 375)
POSTT. 3. Day	0.04 (0.01 / 0.49)	524 (187 / 985)	400.5 (158 /	2.9 (1 / 78)	41.5 (8 / 142)	61.5 (8 / 443)
POSTT. 5. Day	0.03 (0.01 / 0.2)	440 (175 / 1200)	371 (149 / 2197)	3 (1 / 202)	38 (11 / 360)	67 (5 / 480)
P (intragroup)	**<0.001**	**<0.001**	0.929	0.058	**0.048**	**<0.001**
PRET. vs POSTT. 1. Day	**0.003**	**0.049**	ns.	ns.	0.999	0.999
PRET. vs POSTT. 3. Day	**<0.001**	**<0.001**	ns.	ns.	**0.030**	**0.004**
PRET. vs POSTT. 5. Day	**<0.001**	**<0.001**	ns.	ns.	0.035	**<0.001**
POSTT. 1. Day vs POSTT. 3. Day	**<0.001**	**<0.001**	ns.	ns.	0.440	0.247
POSTT. 1. Day vs POSTT. 5. Day	**<0.001**	**<0.001**	ns.	ns.	0.504	**0.007**
POSTT. 3. Day vs POSTT. 5.	**0.121**	**0.001**	ns.	ns.	0.999	0.999

Friedman Test(Monte Carlo); Posthoc Test: Stepwise step-down comparisons),
min: Minimum, max: Maximum, ns.: Not significant, PRET.:Before tocilizumab, POSTT.:After tocilizumab LDH: lactate dehydrogenase AST: aspartate transaminase ALT: alanine transaminase

Table 5. Comparison of Changes in Clinical Findings with Tocilizumab Treatment

	Fever		P	Cough		P	Dyspnea		P	Anorexia		P
	Exitus	Alive		Exitus	Alive		Exitus	Alive		Exitus	Alive	
	n (%)	n (%)		n (%)	n (%)		n (%)	n (%)		n (%)	n (%)	
PRET.	14(100)	62(100)	-	14(100)	62(100)	-	14(100)	61(98.4)	0.999[f]	14(100)	62(100)	-
POSTT. 1. day	14(100)	62(100)	-	14(100)	62(100)	-	14(100)	62(100)	-	14(100)	62(100)	-
POSTT. 3. day	14(100)	28(45.2)	**<0.001[c]**	14(100)	62(100)	-	14(100)	61(98.4)	0.999[f]	14(100)	49(79)	0.110 [f]
POSTT. 5. day	6(42.9)	0(0)	**<0.001[f]**	14(100)	62(100)	-	14(100)	58(93.5)	0.999[f]	13(92.9)	13(21)	**<0.001[f]**
Difference												
(POSTT. 1. day-PRET.)			-			-			0.999[f]			-
Decreased	0(0)	0(0)		0(0)	0(0)		0(0)	0(0)		0(0)	0(0)	
No change	14(100)	62(100)		14(100)	62(100)		14(100)	61(98.4)		14(100)	62(100)	
Increased	0(0)	0(0)		0(0)	0(0)		0(0)	1(1.6)		0(0)	0(0)	
(POSTT. 3. day-PRET.)			**<0.001[c]**			-			0.999 [ff]			0.110[f]
Decreased	0(0)	34(54.8)		0(0)	0(0)		0(0)	1(1.6)		0(0)	13(21)	
No change	14(100)	28(45.2)		14(100)	62(100)		14(100)	60(96.8)		14(100)	49(79)	
Increased	0(0)	0(0)		0(0)	0(0)		0(0)	1(1.6)		0(0)	0(0)	
(POSTT. 5. day-PRET.)			**<0.001[f]**			-			0.999[ff]			**<0.001[f]**
Decreased	8(57.1)	62(100)		0(0)	0(0)		0(0)	4(6.5)		1(7.1)	49(79)	
No change	6(42.9)	0(0)		14(100)	62(100)		14(100)	57(91.9)		13(92.9)	13(21)	
Increased	0(0)	0(0)		0(0)	0(0)		0(0)	1(1.6)		0(0)	0(0)	
(POSTT. 3-1. day)			**<0.001[c]**			-			0.999[f]			0.110[f]
Decreased	0(0)	34(54.8)		0(0)	0(0)		0(0)	1(1.6)		0(0)	13(21)	
No change	14(100)	28(45.2)		14(100)	62(100)		14(100)	61(98.4)		14(100)	49(79)	
Increased	0(0)	0(0)		0(0)	0(0)		0(0)	0(0)		0(0)	0(0)	
(POSTT. 5-1. day)			**<0.001[f]**			-			0.999[f]			**<0.001[f]**
Decreased	8(57.1)	62(100)		0(0)	0(0)		0(0)	4(6.5)		1(7.1)	49(79)	
No change	6(42.9)	0(0)		14(100)	62(100)		14(100)	58(93.5)		13(92.9)	13(21)	
Increased	0(0)	0(0)		0(0)	0(0)		0(0)	0(0)		0(0)	0(0)	
(POSTT. 5-3. day)			0.556 [c]			-			0.999[f]			**0.001[c]**
Decreased	8(57.1)	28(45.2)		0(0)	0(0)		0(0)	3(4.8)		1(7.1)	36(58.1)	
No change	6(42.9)	34(54.8)		14(100)	62(100)		14(100)	59(95.2)		13(92.9)	26(41.9)	
Increased	0(0)	0(0)		0(0)	0(0)		0(0)	0(0)		0(0)	0(0)	

[f] Fisher Exact Test (Monte Carlo), [ff] Fisher Freeman Halton test (Monte Carlo), [c] Pearson Chi Square Test (Monte Carlo),
PRET.:Before tocilizumab, POSTT.:After tocilizumab

Table 6. Comparison of Changes in Laboratory Findings with Tocilizumab Treatment

	Exitus (n = 14)	Alive (n = 62)	P	Exitus (n = 14)	Alive (n = 62)	P
	M.(min/max)	M.(min/max)		M.(min/max)	M.(min/max)	
	SaO2			D-dimer		
PRET.	86.5(85/88)	89(85/92)	**<0.001**	1.65(0.29/5.54)	0.76(0.27/3.99)	**0.034**
POSTT. 1. day	87(86/88)	90(87/92)	**<0.001**	1.74(0.39/20)	0.9(0.27/3.98)	0.095
POSTT. 3. day	87(86/89)	91(88/93)	**<0.001**	1.89(0.4/20)	0.92(0.12/3)	**0.001**
POSTT. 5. day	87(86/89)	93(90/95)	**<0.001**	2.64(0.48/3.87)	0.8(0.1/3.52)	**<0.001**
Difference						
(POSTT.1.day-PRET.)	0(0/1)	1(0/2)	**<0.001**	0.09(-3.63/16.01)	0.06(-2.08/1.69)	0.926
(POSTT.3.day-PRET.)	0.5(-1/2)	3(1/5)	**<0.001**	0.39(-3.51/16.01)	0.07(-3.87/2.25)	0.077
(POSTT.5.day-PRET.)	0(-1/2)	5(1/7)	**<0.001**	0.6(-1.93/2.1)	-0.01(-2.79/1.47)	**0.016**
(POSTT.3-1.day)	0(-1/1)	2(0/3)	**<0.001**	0.18(-0.94/1.03)	0.03(-2.97/2)	0.087
(POSTT.5-1.day)	0(-1/1)	4(1/6)	**<0.001**	0.69(-16.14/2.02)	-0.03(-3.07/1.43)	**0.003**
(POSTT.5-3.day)	0(-2/1)	2(0/4)	**<0.001**	0.55(-16.14/1.8)	-0.07(-2.9/3.4)	**0.008**
	CRP			WBC		
PRET.	138.5(40/252)	121(45/350)	0.598	7460(1950/20240)	8090(2630/19060)	0.558
POSTT. 1. day	78.5(15/171)	82(15/315)	0.865	8790(2620/19450)	8855(3000/17270)	0.864
POSTT. 3. day	27(5/113)	20(1/126)	0.437	9145(2920/21040)	9065(2860/23100)	0.845
POSTT. 5. day	9(1/68)	6(1/63)	0.092	9590(3060/22170)	8960(1060/19720)	0.262
Difference						
(POSTT.1.day-PRET.)	-57(-100/-10)	-48.5(-174/-1)	0.425	925(-3860/3770)	60(-12630/7840)	0.306
(POSTT.3.day-PRET.)	-100(-190/-30)	-96(-309/-24)	0.969	1610(-3070/10240)	995(-12520/9090)	0.544
(POSTT.5.day-PRET.)	-121.5(-234/-37)	-111.5(-337/-42)	0.872	2510(-3750/9130)	1320(-16930/7780)	0.083
(POSTT.3-1.day)	-44.5(-97/-5)	-54(-274/-6)	0.585	930(-3600/7980)	395(-7950/9280)	0.450
(POSTT.5-1.day)	-70.5(-142/-10)	-74(-302/-12)	0.385	2200(-3649/6870)	455(-14010/9080)	0.088
(POSTT.5-3.day)	-7.5(-80/25)	-13(-67/0)	0.168	635(-4719/5850)	280(-13940/6340)	0.346
	Ferritin			PMNL		
PRET.	1007(205/2000)	734.5(83/2000)	0.439	6715(1000/18190)	7235(1450/17420)	0.851
POSTT. 1. day	1034.5(193/2000)	841(93/2000)	0.706	7630(2120/16970)	7580(1900/15950)	0.674
POSTT. 3. day	951.5(209/2000)	729(154/2000)	0.285	8235(2460/19350)	6245(1720/21280)	0.228
POSTT. 5. day	879.5(154/2000)	703(65/1996)	0.451	9000(2710/18360)	7225(2300/16660)	**0.041**
Difference						
(POSTT.1.day-PRET.)	7(-371/474)	58(-439/1332)	0.232	1020(-3920/3510)	-30(-13850/8370)	0.253
(POSTT.3.day-PRET.)	0(-456/991)	0(-973/609)	0.718	1790(-2950/9980)	260(-14010/9400)	0.155
(POSTT.5.day-PRET.)	-100(-668/519)	-23(-1161/916)	0.762	2500(-3690/8910)	85(-14300/7290)	**0.027**
(POSTT.3-1.day)	0(-506/517)	-62(-1057/474)	0.134	1180(-3420/7740)	-175(-9070/9600)	0.199
(POSTT.5-1.day)	-69(-668/421)	-55.5(-1187/677)	0.939	1685(-4620/6670)	-380(-10030/7350)	**0.027**
(POSTT.5-3.day)	-56(-1167/891)	-40(-599/503)	0.578	235(-6130/5400)	-30(-13220/5110)	0.487

Mann Whitney U Test (Monte Carlo)

PRET.:Before tocilizumab, POSTT.:After tocilizumab, M.:Median, min: Minimum, max: Maximum SaO2: oxygen saturation CRP: C-reactive protein WBC: white blood cell PMNL: polymorphonuclear leukocyte

Table 7. Comparison of Changes in Laboratory Findings with Tocilizumab Treatment

	Exitus (n = 14)	Alive (n = 62)	P	Exitus (n = 14)	Alive (n = 62)	P
	M.(min/max)	M.(min/max)		M.(min/max)	M.(min/max)	
	Lymphocyte			**LDH**		
PRET.	505 (220/1130)	660 (180/1600)	0.086	568 (254/995)	362 (132/1077)	**0.001**
POSTT. 1. day	390 (190/1380)	880 (110/2160)	**0.004**	604.5 (291/987)	360 (132/1053)	**0.001**
POSTT. 3. day	465 (160/1680)	1125 (130/3000)	**<0.001**	657.5 (316/1082)	375.5 (158/885)	**<0.001**
POSTT. 5. day	375 (210/2290)	1400 (180/3940)	**<0.001**	807.5 (372/2197)	342.5 (149/1077)	**<0.001**
Difference						
(POSTT. 1.day - PRET.)	40 (-500/540)	145 (-480/1440)	0.085	13.5 (-123/89)	-6 (-222/124)	0.519
(POSTT. 3.day - PRET.)	100 (-620/840)	485 (-330/2100)	**0.001**	63 (-130/462)	-12.5 (-322/237)	**0.014**
(POSTT. 5.day - PRET.)	35 (-600/1160)	694 (-210/2980)	**<0.001**	140.5 (-188/1532)	-18 (-314/572)	**<0.001**
(POSTT. 3-1. day)	40 (-260/300)	180 (-540/1510)	**0.025**	63 (-122/405)	-23 (-335/272)	**0.022**
(POSTT. 5-1. day)	35 (-317/920)	550 (-438/2240)	**<0.001**	188 (-99/1532)	-3 (-383/484)	**0.001**
(POSTT. 5-3. day)	35 (-300/990)	255 (-390/2110)	**0.003**	45.5 (-83/1435)	-11.5 (-193/369)	**0.013**
	Procalcitonin			**Troponin**		
PRET.	0.17 (0.03/1.07)	0.06 (0.02/0.9)	**0.010**	8.5 (1/70)	3 (1/120)	0.056
POSTT. 1. day	0.16 (0.03/0.75)	0.05 (0.01/0.53)	**0.001**	7.5 (1/82)	3 (1/81)	**0.019**
POSTT. 3. day	0.07 (0.03/0.49)	0.03 (0.01/0.22)	**<0.001**	8 (1/69)	2 (1/78)	**0.010**
POSTT. 5. day	0.05 (0.01/0.2)	0.03 (0.01/0.15)	**0.002**	6.5 (1/202)	3 (1/69)	0.042
Difference						
(POSTT. 1.day - PRET.)	-0.05 (-0.61/0.23)	-0.01 (-0.54/0.02)	0.676	-2 (-23/52)	0 (-51/42)	0.565
(POSTT. 3.day - PRET.)	-0.09 (-0.76/0.02)	-0.03 (-0.8/0.01)	**0.029**	0 (-31/38)	0 (-98/39)	0.400
(POSTT. 5.day - PRET.)	-0.13 (-0.92/0.02)	-0.03 (-0.86/0.02)	**0.036**	-1.5 (-52/164)	0 (-103/25)	0.879
(POSTT. 3-1. day)	-0.07 (-0.43/0.02)	-0.01 (-0.43/0.02)	**0.013**	-1.5 (-18/39)	0 (-47/56)	0.819
(POSTT. 5-1. day)	-0.14 (-0.55/0.01)	-0.02 (-0.45/0.02)	**0.014**	-1 (-67/172)	-0.5 (-52/58)	0.822
(POSTT. 5-3. day)	-0.02 (-0.3/0.01)	-0.01 (-0.12/0.04)	0.072	-2 (-53/133)	0 (-22/9)	0.267
	ALT			**AST**		
PRET.	37 (8/100)	36.5 (9/308)	0.517	44 (17/84)	33.5 (8/152)	0.409
POSTT. 1. day	35.5 (11/127)	46.5 (8/375)	0.225	34.5 (14/102)	36 (8/177)	0.754
POSTT. 3. day	39.5 (8/192)	64 (8/443)	0.178	34 (15/103)	42 (8/142)	0.416
POSTT. 5. day	34 (5/339)	69 (7/480)	**0.046**	35 (17/360)	39 (11/136)	0.473
Difference						
(POSTT. 1.day - PRET.)	-0.5 (-14/27)	3 (-32/106)	0.138	-4 (-27/18)	1.5 (-39/72)	**0.027**
(POSTT. 3.day - PRET.)	3 (-18/134)	17 (-28/197)	0.112	-1 (-37/49)	6.5 (-95/97)	0.205
(POSTT. 5.day - PRET.)	1.5 (-28/281)	22.5 (-17/213)	**0.028**	1 (-48/293)	2 (-95/80)	0.661
(POSTT. 3-1. day)	-2.5 (-13/137)	9.5 (-68/91)	0.358	-4 (-27/18)	1.5 (-39/72)	**0.027**
(POSTT. 5-1. day)	-1.5 (-54/284)	19.5 (-72/116)	0.092	5 (-65/299)	3.5 (-123/72)	0.911
(POSTT. 5-3. day)	-3 (-48/240)	6 (-61/92)	0.121	0 (-45/257)	0.5 (-93/50)	0.941

Mann Whitney U Test (Monte Carlo)

PRET.:Before tocilizumab, POSTT.:After tocilizumab, M.:Median, min: Minimum, max: Maximum, ALT: alanine transaminase AST: aspartate transaminase

Table 8. Comparison of Changes in Laboratory Findings with Tocilizumab Treatment

	Exitus (n = 14)	Alive (n = 62)	p value
	Median (min/max)	Median (min/max)	
Fibrinogen			
PRET.	665.5 (383/921)	713.5 (402/1200)	0.459
POSTT. 1. day	623.5 (331/939)	659.5 (309/1026)	0.370
POSTT. 3. day	524.5 (236/846)	524 (187/985)	0.615
POSTT. 5. day	428 (175/711)	440 (216/1200)	0.806
Difference			
(POSTT. 1.day - PRET.)	-67.5 (-226/129)	-75 (-234/207)	0.869
(POSTT. 3.day - PRET.)	-174 (-303/99)	-176 (-534/218)	0.792
(POSTT. 5.day - PRET.)	-259 (-594/209)	-265 (-788/348)	0.762
(POSTT. 3-1. day)	-91.5 (-246/80)	-110 (-458/170)	0.475
(POSTT. 5-1. day)	-178 (-492/116)	-229.5 (-579/309)	0.504
(POSTT. 5-3. day)	-77 (-402/278)	-73.5 (-396/215)	0.912

Mann Whitney U Test (Monte Carlo),

PRET.:Before tocilizumab, POSTT.:After tocilizumab, min: Minimum, max: Maximum

Table 9. ROC Analysis Indicating the Clinical and Laboratory Findings Associated with Mortality

Dependent Variable: Mortality	Cut off	Sensitivity	Specificity	+PV	-PV	AUC±SE.	p value
SaO2 (Difference)							
(POSTT. 1.day - PRET.)	≤ 0	64.29%	88.71%	56.2	91.7	0.794 ± 0.070	**<0.001**
(POSTT. 3.day - PRET.)	≤ 1	92.86%	88.71%	65.0	98.2	**0.958 ± 0.023**	**<0.001**
(POSTT. 5.day - PRET.)	≤ 2	100.00%	96.77%	87.5	100.0	**0.995 ± 0.005**	**<0.001**
(POSTT. 3-1. day)	≤ 0	64.29%	96.77%	81.8	92.3	**0.912 ± 0.038**	**<0.001**
(POSTT. 5-1. day)	≤ 1	100.00%	96.77%	87.5	100.0	**0.994 ± 0.006**	**<0.001**
(POSTT. 5-3. day)	≤ 1	100.00%	85.48%	60.9	100.0	**0.979 ± 0.014**	**<0.001**
D-dimer (Difference)							
(POSTT. 3 - PRET.)	> 0.34	57.14%	80.65%	40.0	89.3	0.654 ± 0.098	0.115
(POSTT. 5 - PRET.)	> 0.18	71.43%	77.42%	41.7	92.3	0.707 ± 0.096	**0.031**
(POSTT. 3-1. day)	> 0.22	50.00%	80.65%	36.8	87.7	0.648 ± 0.086	0.085
(POSTT. 5-1. day)	> 0.44	57.14%	88.71%	53.3	90.2	0.747 ± 0.087	**0.005**
(POSTT. 5-3. day)	> 0.06	78.57%	75.81%	42.3	94.0	0.731 ± 0.097	**0.017**
WBC (Difference)							
(POSTT. 5.day - PRET.)	> 510	85.71%	45.16%	26.1	93.3	0.647 ± 0.077	0.057
(POSTT. 5-1. day)	> 1730	64.29%	70.97%	33.3	89.8	0.648 ± 0.079	0.061
PMNL (Difference)							
(POSTT. 5.day - PRET.)	> 2400	57.14%	75.81%	34.8	88.7	0.690 ± 0.080	**0.019**
(POSTT. 5-1. day)	> 150	85.71%	58.06%	31.6	94.7	0.690 ± 0.079	**0.016**
Lymphocyte (Difference)							
(POSTT. 3.day - PRET.)	≤ 300	92.86%	64.52%	37.1	97.6	0.775 ± 0.063	**<0.001**
(POSTT. 5.day - PRET.)	≤ 170	71.43%	88.71%	58.8	93.2	**0.831 ± 0.064**	**<0.001**
(POSTT. 3-1. day)	≤ 300	100.00%	41.94%	28.0	100.0	0.694 ± 0.067	**0.004**
(POSTT. 5-1. day)	≤ 370	92.86%	66.13%	38.2	97.6	0.796 ± 0.060	**<0.001**
(POSTT. 5-3. day)	≤ 80	85.71%	69.35%	38.7	95.6	0.757 ± 0.079	**0.001**
Procalcitonin (Difference)							
(POSTT. 3.day - PRET.)	≤ -0.04	78.57%	62.90%	32.4	92.9	0.688 ± 0.090	**0.036**
(POSTT. 5.day - PRET.)	≤ -0.11	64.29%	79.03%	40.9	90.7	0.680 ± 0.092	**0.050**
(POSTT. 3-1. day)	≤ -0.03	78.57%	70.97%	35.5	93.3	0.704 ± 0.089	**0.022**
(POSTT. 5-1. day)	≤ -0.04	78.57%	70.97%	35.7	91.7	0.708 ± 0.096	**0.030**
(POSTT. 5-3. day)	≤ -0.03	50.00%	83.87%	45.5	86.2	0.657 ± 0.093	**0.091**
LDH (Difference)							
(POSTT. 3.day - PRET.)	> 39	64.29%	79.03%	40.9	90.7	0.710 ± 0.074	**0.004**
(POSTT. 5.day - PRET.)	> 90	78.57%	88.71%	61.1	94.8	**0.809 ± 0.074**	**<0.001**
(POSTT. 3-1. day)	> 0	85.71%	59.68%	32.4	94.9	0.693 ± 0.080	**0.016**
(POSTT. 5-1. day)	> 80	71.43%	83.87%	50.0	92.9	0.767 ± 0.079	**0.001**

(POSTT. 5-3. day)	> 14	78.57%	66.13%	34.4	93.2	0.713 ± 0.073	**0.004**
AST (Difference)							
(POSTT. 1.day - PRET.)	≤ 3	92.86%	45.16%	27.7	96.6	0.692 ± 0.072	**0.007**
(POSTT. 3-1. day)	≤ 3	92.86%	45.16%	27.7	96.6	0.692 ± 0.075	**0.007**
ALT (Difference)							
(POSTT. 5.day - PRET.)	≤ 11	78.57%	64.52%	33.3	93.0	0.686 ± 0.090	**0.038**
(POSTT. 5-1. day)	≤ 22	85.71%	48.39%	27.3	93.7	0.645 ± 0.087	0.098

ROC (Receiver Operating Curve) Analysis (Honley&Mc Nell - Youden indExitus J), AUC: Area under the ROC curve, SE: Standard Error, PRET.:Before tocilizumab,POSTT.:After tocilizumab,
+PV: Positive Predictive Value, -PV: Negative Predictive Value SaO2: oxygen saturation WBC: white blood cell PMNL: polymorphonuclear leukocyte LDH: lactate dehydrogenase AST: aspartate transaminase ALT: alanine transaminase

Table 10. Kaplan-Meier Analysis Indicating the Factors Associated with Mortality

		Exitus	Alive	Estimate Survival	Estimate Proportion Surviving at the	P value
		n(%)	n(%)	Mean ± Se.	10 day/20 day/30 day	
Fever (Difference)						
(POSTT. 3. day - PRET.)	Decreased	0 (0)	34 (54.8)	-	100/100/100	**0.015**
	No change	14 (100)	28 (45.2)	-	95.1/68.3/37.4	
(POSTT. 5. day - PRET.)	Decreased	8 (57.1)	62 (100)	34.882±3.084	98.4/88.2/55.2	**0.001**
	No change	6 (42.9)	0 (0)	20.833±4.729	83.3/33.3/16.7	
(POSTT. 3-1. day)	Decreased	0 (0)	34 (54.8)	-	100/100/100	**0.015**
	No change	14 (100)	28 (45.2)	-	95.1/68.3/37.4	
(POSTT. 5-1. day)	Decreased	8 (57.1)	62 (100)	34.882±3.084	98.4/88.2/55.2	**0.001**
	No change	6 (42.9)	0 (0)	20.833±4.729	83.3/33.3/16.7	
Anorexia (Difference)						
(POSTT. 5. day - PRET.)	Resolved	1 (7.1)	49 (79)	34.500±3.182	100/100/50	**0.002**
	No change	13 (92.9)	13 (21)	27.080±3.114	92.3/56.7/37.8	
(POSTT. 5-1. day)	Resolved	1 (7.1)	49 (79)	34.500±3.182	100/100/50	**0.002**
	No change	13 (92.9)	13 (21)	27.080±3.114	92.3/56.7/37.8	
(POSTT. 5-3. day)	Resolved	1 (7.1)	36 (58.1)	30.000±0.000	100/100/0	**0.047**
	No change	13 (92.9)	26 (41.9)	30.036±2.816	94.7/67.7/48.4	
SaO2 (Difference)						

(POSTT. 1. day - PRET.)	> 0	0 (0)	10 (16.1)	-	100/100/100	0.267
	≤ 0	14 (100)	52 (83.9)	-	96.7/76.5/46.6	
(POSTT. 3. day - PRET.)	> 1	0 (0)	38 (61.3)	-	100/100/100	**0.005**
	≤ 1	14 (100)	24 (38.7)	-	94.5/65.1/35.6	
(POSTT. 5. day - PRET.)	> 2	0 (0)	56 (90.3)	-	100/100/100	**<0.001**
	≤ 2	14 (100)	6 (9.7)	-	90.0/47.9/21.3	
(POSTT. 3-1. day)	> 0	0 (0)	36 (58.1)	-	100/100/100	**0.009**
	≤ 0	14 (100)	26 (41.9)	-	94.8/66.4/36.3	
(POSTT. 5-1. day)	> 1	0 (0)	57 (91.9)	-	100/100/100	**<0.001**
	≤ 1	14 (100)	5 (8.1)	-	89.5/46.8/20.8	
(POSTT. 5-3. day)	> 1	0 (0)	16 (25.8)	-	100/100/100	0.177
	≤ 1	14 (100)	46 (74.2)	-	96.5/75.8/45.5	
D-dimer (Difference)						
(POSTT. 5. day - PRET.)	≤ 0.18	4 (28.6)	47 (75.8)	33.093±2.593	100/82.4/65.9	0.100
	> 0.18	10 (71.4)	15 (24.2)	28.563±3.402	91.1/70.1/35.1	
(POSTT. 3-1. day)	≤ 0.22	7 (50)	49 (79)	34.860±3.354	100/79.8/63.8	0.231
	> 0.22	7 (50)	13 (21)	27.991±3.555	89.2/76.0/25.3	
(POSTT. 5-1. day)	≤ 0.44	6 (42.9)	52 (83.9)	37.232±2.533	98.1/92.0/78.9	**0.005**
	> 0.44	8 (57.1)	10 (16.1)	23.373±2.047	94.1/60.1/0.0	
(POSTT. 5-3. day)	≤ 0.06	3 (21.4)	46 (74.2)	36.747±2.895	97.8/97.8/78.2	**0.011**
	> 0.06	11 (78.6)	16 (25.8)	26.511±3.387	96.2/55.4/27.7	
WBC (Difference)						
(POSTT. 5. day - PRET.)	≤ 510	2 (14.3)	27 (43.5)	35.750±2.099	100/83.3/83.3	0.103
(POSTT. 5-1. day)	≤ 1730	5 (35.7)	43 (69.4)	32.783±3.028	100/88.5/88.5	0.067
	> 1730	9 (64.3)	19 (30.6)	30.084±3.908	92.9/66.7/50.0	

Kaplan Meier Test; Log Rank (Mantel-Cox), SE.: Standard Error, PRET.:Before tocilizumab,POSTT.:After tocilizumab SaO2: oxygen saturation WBC: white blood cell

Table 11. Kaplan-Meier Analysis Indicating the Factors Associated with Mortality

		Exitus	Alive	Estimate Survival	Estimate Proportion Surviving at the	P value
		n(%)	n(%)	Mean ± SE.	10 day/20 day/30 day	
PMNL (Difference)						
(POSTT. 5. day - PRET.)	≤ 2400	6 (42.9)	46 (74.2)	35.567±2.814	100/89.5/89.5	**0.001**
	> 2400	8 (57.1)	16 (25.8)	21.918±1.986	91.3/61.6/30.8	
(POSTT. 5-1. day)	≤ 150	2 (14.3)	35 (56.5)	38.500±2.044	100/92.9/92.9	**0.022**
	> 150	12 (85.7)	27 (43.5)	28.096±3.134	94.7/67.0/29.8/	
Lymphocyte (Difference)						
(POSTT. 3. day - PRET.)	> 300	1 (7.1)	17 (27.4)	43.000±0.000	100/100/0	0.054
	≤ 300	13 (92.9)	45 (72.6)	29.676±2.372	96.4/74.6/40.8	
(POSTT. 5. day - PRET.)	> 170	4 (28.6)	55 (88.7)	38.411±2.615	100/95.8/82.1	**<0.001**
	≤ 170	10 (71.4)	7 (11.3)	20.729±2.258	87.8/46.2/0	
(POSTT. 3-1. day)	> 300	0 (0)	26 (41.9)	-	100/100/100	**0.028**
	≤ 300	14 (100)	36 (58.1)	-	95.8/71.3/36.7	
(POSTT. 5-1. day)	> 370	1 (7.1)	41 (66.1)	33.750±1.949	100/100/75.0	**0.010**
	≤ 370	13 (92.9)	21 (33.9)	28.395±2.967	93.8/63.4/38.0	
(POSTT. 5-3. day)	> 80	2 (14.3)	43 (69.4)	37.833±2.797	100/100/83.3	**0.004**
	≤ 80	12 (85.7)	19 (30.6)	25.930±3.236	93/60.2/20.1	
Procalcitonin (Difference)						
(POSTT. 3. day - PRET.)	> -0.04	3 (21.4)	39 (62.9)	33.523±2.533	100/95.5/53	**0.040**
	≤ -0.04	11 (78.6)	23 (37.1)	28.909±3.532	93.9/63.5/42.3	
(POSTT. 5. day - PRET.)	> -0.11	5 (35.7)	48 (77.4)	32.579±2.500	97.9/91.7/50.9	0.148
	≤ -0.11	9 (64.3)	14 (22.6)	28.935±3.774	95.7/62.0/41.3	
(POSTT. 3-1. day)	> -0.03	3 (21.4)	42 (67.7)	32.973±3.154	97.4/93.4/46.7	0.080
	≤ -0.03	11 (78.6)	20 (32.3)	28.907±3.187	96.8/66.3/39.8	
(POSTT. 5-1. day)	> -0.04	3 (21.4)	44 (71)	33.071±3.145	97.6/93.8/46.9	0.051
	≤ -0.04	11 (78.6)	18 (29)	28.435±3.226	96.6/64.0/38.4	
(POSTT. 5-3. day)	> -0.03	7 (50)	52 (83.9)	31.934±4.022	98.1/86.1/34.4	0.194
	≤ -0.03	7 (50)	10 (16.1)	28.983±3.674	94.1/64.9/48.7	
LDH (Difference)						
(POSTT. 3. day - PRET.)	≤ 39	5 (35.7)	48 (77.4)	29.331±2.483	100/76.0/50.7	0.425
	> 39	9 (64.3)	14 (22.6)	30.723±3.376	91.3/76.5/47.8	
(POSTT. 5. day - PRET.)	≤ 90	3 (21.4)	54 (87.1)	40.063±2.380	100/87.5/87.5	**<0.001**
	> 90	11 (78.6)	8 (12.9)	23.505±3.069	89.5/57.6/17.3	
(POSTT. 3-1. day)	≤ 0	2 (14.3)	35 (56.5)	25.317±1.092	100/81.7/81.7	0.267
	> 0	12 (85.7)	27 (43.5)	30.867±2.735	92.2/76.4/45.8	
(POSTT. 5-1. day)	≤ 80	4 (28.6)	51 (82.3)	39.222±2.372	100/84.7/84.7	**0.001**
	> 80	10 (71.4)	11 (17.7)	24.452±3.181	90.0/62.7/18.8	
(POSTT. 5-3. day)	≤ 14	3 (21.4)	40 (64.5)	40.398±2.235	100/90.0/90.0	**0.002**
	> 14	11 (78.6)	22 (35.5)	25.174±2.840	89.5/61.7/18.5	
AST (Difference)						
(POSTT. 1. day - PRET.)	> 3	1 (7.1)	27 (43.5)	28.000±0.000	100/100/0	0.127
	≤ 3	13 (92.9)	35 (56.5)	30.787±2.723	95.6/70/49	
(POSTT. 3-1. day)	> 3	1 (7.1)	27 (43.5)	28.000±0.000	100/100/0	0.127
	≤ 3	13 (92.9)	35 (56.5)	30.787±2.723	95.6/70.0/49.0	
ALT (Difference)						
(POSTT. 5. day - PRET.)	> 11	3 (21.4)	38 (61.3)	26.284±0.957	94.7/90.4/90.4	0.416
	≤ 11	11 (78.6)	24 (38.7)	30.520±2.802	100/72.5/42.3	
(POSTT. 5-1. day)	> 22	2 (14.3)	30 (48.4)	26.495±1.033	96.8/91.4/91.4	0.356
	≤ 22	12 (85.7)	32 (51.6)	30.685±2.693	97.5/74.0/43.2	
Co-existent illnesses						0.057
Absent		1 (7.1)	19 (30.6)	40.000±0.000	100/100/100	
Present		13 (92.9)	43 (69.4)	28.153±3.025	96.3/91.17/86.2	

Kaplan Meier Test ; Log Rank (Mantel-Cox), SE.: Standard Error, PRET.:Before tocilizumab,POSTT.:After tocilizumab PMNL: polymorphonuclear leukocyte LDH: lactate dehydrogenase AST: aspartate transaminase ALT: alanine transaminase

Table 12. Cox Regression Analysis Indicating Predictors of Mortality

Independent Variables	B±Sh	p value	Odds Ratio (95%C.I.)
Fever - Difference (POSTT. 5. day - PRET.)	-2.739±0.964	**0,004**	0.065 [0.01-0.428]
Anorexia - Difference (POSTT. 5.day - PRET.)	-0.826±1.209	0,494	0.438 [0.041-4.679]
D-dimer - Difference (POSTT. 5-1. day)	-2.015±0.888	**0,023**	0.133 [0.023-0.759]
WBC - Difference (POSTT. 5-1. day)	-1.772±0.852	**0,037**	0.17 [0.032-0.902]
PMNL - Difference (POSTT. 5. day - PRET.)	-2.583±1.082	**0,017**	0.076 [0.009-0.631]
Procalcitonin - Difference (POSTT. 3. day - PRET.)	-0.256±0.889	0,774	0.774 [0.136-4.42]
LDH - Difference (POSTT. 3-1. day)	0.277±0.948	0,770	1.319 [0.206-8.45]
10 / 20 / 30 day Survival ratios: 99.9 (0.001) / 95.1 (0.032) / 66.0 (0.180) - **Baseline Hazard: 0.742**			

Cox Regression-Stepwise (Wald) Model, B: Regression coefficients, SE: Standard error, C.I.: Confidence interval, PRET.:Before tocilizumab,POSTT.:After tocilizumab WBC: white blood cell PMNL: polymorphonuclear leukocyte LDH: lactate dehydrogenase

DISCUSSION

About one fifth of the patients treated with tocilizumab had died. With tocilizumab therapy, persistence of fever, and an increase in WBC, PMNL and D-dimer levels predicted mortality. An increase in SaO2 and lymphocyte levels was associated with lower mortality rates.

Proinflammatory cytokines were shown to be elevated in COVID-19 infection, and administration of glucocorticoids was observed to decrease the mortality rate by modulating host immune responses (4, 15-21, 26-28). Increased IL-6 levels, which were shown to be associated with mortality, have been a therapeutic target for COVID-19 infection (4, 21). Tocilizumab was shown to decrease mortality in many studies (4,22-24). In a phase 2 study, tocilizumab, when added to standard care decreased the risk of mortality (12 vs. 19%), duration of intensive care admission as well as mechanical ventilation in hospitalized patients with COVID-19 infection (4). In a large meta-analysis, tocilizumab was shown to decrease all-cause mortality, the need for mechanical ventilation and the duration of hospital stays in hospitalized patients with COVID-19 infection (23). Another meta-analysis revealed that those patients under oxygen or noninvasive ventilation support might derive some benefit with respect to mortality risk from tocilizumab use (29). Similar results were also obtained from other meta-analyses (30, 31). The mortality rate in our study was similar to the short-term mortality rate found in some studies, and it was lower or higher than that in other studies (30-33). However, in some studies, the reduction in the risk of mortality was not significant (23, 32-34). Tocilizumab has taken its place in the guidelines regarding the management of hospitalized patients with COVID-19 infection (35, 36).

We showed that persistence of fever or anorexia, increase in SaO2 or lymphocyte count, increase in LDH, PMNL or D-dimer levels, and decrease in procalcitonin levels were associated with mortality. In one study analyzing 106 patients with COVID-19 infection treated with tocilizumab, the mortality rate was 23.6%, and associated with higher IL-6, ferritin, and LDH levels and lower lymphocyte counts (37). In that study, chronic heart and renal disease were also found to be associated with mortality (37). However, we did not show any association of comorbid illnesses with mortality. In another study analyzing 13 severe to critically ill patients with COVID-19 infection treated with tocilizumab, a higher ferritin/CRP ratio was associated with unresponsiveness to treatment (38). Changes in CRP and ferritin levels were similar in exitus and alive groups in our study, and none of these changes predicted mortality. We did not evaluate the ferritin/CRP ratio. In one study, persistently high levels of ferritin after treatment with tocilizumab were shown to indicate mortality (39). The previous studies referred to above did not analyze the association of the changes in inflammatory parameters with mortality.

To our knowledge, those studies investigating the changes in inflammatory parameters in COVID-19 infection treated with tocilizumab are limited. A fall in CRP at 48 hours to a level that was half of that seen at the baseline was shown to predict

overall response to tocilizumab in a small study including patients with COVID-19 infection (40). In contrast to our findings, an increase in D-dimer levels was not associated with non-responders in that study. We showed that ferritin levels increased first after tocilizumab treatment, then returned to pre-treatment levels. A small study suggested that an increase in serum iron and transferrin saturation together with a decrease in ferritin levels might potentiate viral replication in COVID-19 patients (41). In another study, ferritin and IL-6 levels were shown not to change significantly, despite decreases in LDH and CRP levels in those patients with COVID-19 infection under tocilizumab treatment (42). Şener et al. showed that higher CRP, WBC and neutrophil levels, and lower lymphocyte levels on the 1st, 3rd and 5th days after tocilizumab treatment were associated with mortality in those patients with COVID-19 infection (39). But the mortality rate was very high (about 60%), and they did not analyze the difference between pretreatment and posttreatment levels of inflammatory markers. In a study including a relatively larger sample, increasing LDH during treatment and advanced age were predictors for mortality in tocilizumab-treated patients with severe COVID-19 infection (43). Besides fever and SaO2, we recommend the measurement of leukocyte, neutrophil, lymphocyte and D-dimer levels during the course of COVID-19 infection.

Some patients in that study were fully vaccinated. We included those patients, who were infected and followed-up before anti SARS-CoV-2 vaccines had been developed. Therefore, we could not analyze the association of changes in clinical and inflammatory parameters with vaccination against COVID-19 infection.

We initiated tocilizumab treatment in those patients with COVID-19 infection, who were unresponsive to glucocorticoids, or where the clinical course had progressed rapidly. We did not compare their treatment with tocilizumab with any other treatment options; instead, we analyzed the factors associated with mortality in those patients treated with tocilizumab. In the literature, tocilizumab treatment was administered in those patients with COVID-19 infection usually together with glucocorticoids (29, 44).

Elevated liver enzymes were reported after tocilizumab treatment in rheumatic diseases before the COVID-19 pandemic, and in COVID-19 infection (45,46). We did not give tocilizumab to those patients where liver enzymes were elevated ≥5-fold. ALT and AST levels were lower in the exitus group, but higher in the alive group. Changes in ALT or AST levels did not predict mortality. Therefore, the increase in ALT and AST levels might be a side effect of tocilizumab, and the decrease in ALT and AST levels in the exitus group might be the result of decreased liver function.

The risk of neutropenia was found to be higher, but that for thrombosis or bacteremia was not, with tocilizumab treatment in COVID-19 infection (23). Neutropenia was also reported in previous studies analyzing tocilizumab use in rheumatoid arthritis or COVID-19 infection (45, 46). Higher levels of neutrophil or leukocyte predicted mortality in our study. It may be the result of progressive inflammation in exitus patients, and the fact that we included those patients treated with tocilizumab. Moreover, some patients were treated with glucocorticoids together with tocilizumab. Therefore, it was difficult to observe the strict side effects of tocilizumab alone in those patients with COVID-19 infection.

Persistence of fever predicted mortality in our study. Fever is an important finding indicating inflammation in COVID-19 infection. During treatment with tocilizumab, not only inflammatory biomarkers but also symptoms and physical signs should be followed-up to evaluate the progress of the infection.

Strengths and Limitations

We included a considerable number of patients with COVID-19 infection treated with tocilizumab. We measured and followed-up the inflammatory markers before and after tocilizumab treatment, and analyzed the changes. We did not compare these markers and changes with those in patients undergoing other treatment options. We included patients before vaccination against COVID-19 infection had been initiated. Some patients were unresponsive to glucocorticoid treatment, so we could not evaluate the effect of tocilizumab independent of glucocorticoids.

Conclusion

The mortality rate of those patients with COVID-19 infection treated with tocilizumab was low.

Persistence of fever, and increases in leukocyte, neutrophil or D-dimer levels predicted mortality. Studies analyzing the changes in inflammatory markers associated with tocilizumab in COVID-19 infection are limited. We recommend that the changes in the levels of inflammatory markers, especially leukocyte, neutrophil, lymphocyte and D-dimer levels, besides the clinical findings such as fever or SaO2, should be evaluated during the treatment of COVID-19 infection. Future studies, including those of patients vaccinated against COVID-19 infection, and those receiving other treatment options, should reveal the importance of changes in inflammatory markers in these populations also.

Ethics Committee Approval: This retrospective study was conducted in SANKO University Medical Faculty hospital, and approved by the local SANKO University Clinical Research Ethics Committee with approval number of 04 (session no: 2021/06).

Authors' Contributions: All authors contributed to the study conception and design. Study design, material preparation, data collection and analysis were performed by Mustafa Tanrıverdi and Nevhiz Gündoğdu. The first draft of the manuscript was written by first author and co-author commented on previous versions of the manuscript. All authors read and approved the final manuscript.

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

Funding: No funding was taken during the whole process of study, writing of the article or preparation of manuscript.

Acknowledgment: We thank to all authors for major contribution to the study conception, design, data collection and writing of the manuscript.

Informed Consent: Written informed consent was given by all the participants included in the study.

REFERENCES

1- Available from: https://covid19.who.int/

2- Bottle A, Faitna P, Brett S, Aylin P. Factors associated with, and variations in, COVID-19 hospital death rates in England's first two waves: observational study. BMJ Open 2022;12:e060251. doi:10.1136/ bmjopen-2021-060251.

3- Available from: https://covid19.saglik.gov.tr/

4- Rutgers A, Westerweel PE, van der Holt B, Postma S, van Vonderen MGA, Piersma DP, et al. Timely administration of tocilizumab improves outcome of hospitalized COVID-19 patients. PLoS One. 2022 Aug 12;17(8):e0271807. doi: 10.1371/journal.pone.0271807.

5- Bajpai J, Kant S, Verma A, Patwa AK, Atam V, Chaudhary SC, et al. The Severity of COVID 19 Pneumonia in Vaccinated vs. Non-vaccinated Patients in the Second Wave: An Experience From a Tertiary Care Center in India. Cureus 14(5):e25378. doi: 10.7759/cureus.25378.

6- Sonmez A, Demirci I, Haymana C, Tasci I, Dagdelen S, Salman S, et al. Clinical characteristics and outcomes of COVID-19 in patients with type 2 diabetes in Turkey: A nationwide study (TurCoviDia). J Diabetes. 2021;13:585-595.

7- Sahin I, Haymana C, Demir T, Demirci I, Tasci I, Atmaca A, et al. Clinical Characteristics and Outcomes of COVID-19 Patients with Overweight and Obesity: Turkish Nationwide Cohort Study (TurCObesity). Exp Clin Endocrinol Diabetes. 2021. doi: 10.1055/a-1552-4449. Forthcoming.

8- Fattore GL, Olivos NSA, Olalla JEC, Gomez L, Marucco AF, Mena MPR. Mortality in patients with cancer and SARS-CoV-2 infection: Results from the Argentinean Network of Hospital-Based Cancer Registries. Cancer Epidemiol. 2022 Jun 8;79:102200. doi: 10.1016/j.canep.2022.102200.

9- Watson OJ, Barnsley G, Toor J, Hogan AB, Winskill P, Ghani AC. Global impact of the first year of COVID-19 vaccination: a mathematical modelling study. Lancet Infect Dis. 2022 Jun 23:S1473-3099(22)00320-6. doi: 10.1016/S1473-3099(22)00320-6.

10- Wells CR, Galvani AP. The global impact of disproportionate vaccination coverage on COVID-19 mortality. Lancet Infect Dis. 2022 Jun 23:S1473-3099(22)00417-0. doi: 10.1016/S1473-3099(22)00417-0.

11- Wong CKH, Lau KTK, Xiong X, Au ICH, Lai FTT, Wan EYF, et al. Adverse events of special interest and mortality following vaccination with mRNA (BNT162b2) and

inactivated (CoronaVac) SARS-CoV-2 vaccines in Hong Kong: A retrospective study. PLoS Med. 2022 Jun 21;19(6):e1004018. doi: 10.1371/journal.pmed.1004018.

12- Lopez Bernal J, Andrews N, Gower C, Robertson C, Stowe J, Tessier E, et al. Effectiveness of the Pfizer-BioNTech and Oxford-AstraZeneca vaccines on covid-19 related symptoms, hospital admissions, and mortality in older adults in England: test negative case-control study. BMJ. 2021 May 13;373:n1088. doi: 10.1136/bmj.n1088.

13- Wiersinga WJ, Rhodes A, Cheng AC, Peacock SJ, Prescott HC. Pathophysiology, Transmission, Diagnosis, and Treatment of Coronavirus Disease 2019 (COVID-19): A Review. JAMA. 2020 Aug 25;324(8):782-793. doi: 10.1001/jama.2020.12839.

14- Choudhary S, Sharma K, Silakari O. The interplay between inflammatory pathways and COVID-19: A critical review on pathogenesis and therapeutic options. Microb Pathog. 2021 Jan;150:104673. doi: 10.1016/j.micpath.2020.104673.

15- Topcu H, Arik YE. The Importance of D-Dimer, Ferritin, CRP and Lymphocyte Values in Determining Mortality in COVID-19 Disease in Turkey. Clin Lab. 2022 May 1;68(5). doi: 10.7754/Clin.Lab.2021.210720.

16- Tjendra Y, Al Mana AF, Espejo AP, Akgun Y, Millan NC, Gomez-Fernandez C, et al. Predicting Disease Severity and Outcome in COVID-19 Patients: A Review of Multiple Biomarkers. Arch Pathol Lab Med. 2020 Dec 1;144(12):1465-1474. doi: 10.5858/arpa.2020-0471-SA.

17- Broman N, Rantasärkkä K, Feuth T, Valtonen M, Waris M, Hohenthal U, et al. IL-6 and other biomarkers as predictors of severity in COVID-19. Ann Med. 2021 Dec;53(1):410-412. doi: 10.1080/07853890.2020.1840621.

18- Huang I, Pranata R, Lim MA, Oehadian A, Alisjahbana B. C-reactive protein, procalcitonin, D-dimer, and ferritin in severe coronavirus disease-2019: a meta-analysis. Ther Adv Respir Dis. 2020 Jan-Dec;14:1753466620937175. doi: 10.1177/1753466620937175.

19- Sanchez BG, Gasalla JM, Sánchez-Chapado M, Bort A, Diaz-Laviada I. Increase in Ischemia-Modified Albumin and Pregnancy-Associated Plasma Protein-A in COVID-19 Patients. J Clin Med. 2021 Nov 23;10(23):5474. doi: 10.3390/jcm10235474.

20- La Torre G, Marte M, Massetti AP, Carli SM, Romano F, Mastroianni CM, et al. The neutrophil/lymphocyte ratio as a prognostic factor in COVID-19 patients: a case-control study. Eur Rev Med Pharmacol Sci. 2022 Feb;26(3):1056-1064. doi: 10.26355/eurrev_202202_28017.

21- Del Valle DM, Kim-Schulze S, Huang HH, Beckmann ND, Nirenberg S, Wang B, et al. An inflammatory cytokine signature predicts COVID-19 severity and survival. Nat Med. 2020 Oct;26(10):1636-1643. doi: 10.1038/s41591-020-1051-9.

22- Oliynyk O, Barg W, Oliynyk Y, Dubrov S, Gurianov V, Rorat M. Lack of Difference in Tocilizumab Efficacy in the Treatment of Severe COVID-19 Caused by Different SARS-CoV-2 Variants. J Pers Med. 2022 Jul 4;12(7):1103. doi: 10.3390/jpm12071103.

23- Piscoya A, Parra Del Riego A, Cerna-Viacava R, Rocco J, Roman YM, Escobedo AA, et al. Efficacy and harms of tocilizumab for the treatment of COVID-19 patients: A systematic review and meta-analysis. PLoS One. 2022 Jun 3;17(6):e0269368. doi: 10.1371/journal.pone.0269368.

24- Alex R, Gulam SM, Kumar K. Real-Life Use of Tocilizumab in the Treatment of Severe COVID-19 Pneumonia. Adv Virol. 2022 Jun 9;2022:7060466. doi: 10.1155/2022/7060466.

25- Available from: https://covid19.saglik.gov.tr/TR-66341/antisitokin-antiinflamatuar-tedaviler-koagulopati-yonetimi.html

26- Wang D, Hu B, Hu C, Zhu F, Liu X, Zhang J, et al. Clinical Characteristics of 138 Hospitalized Patients With 2019 Novel Coronavirus-Infected Pneumonia in Wuhan, China. JAMA. 2020 Mar 17;323(11):1061-1069. doi: 10.1001/jama.2020.1585.

27- Zhu Z, Cai T, Fan L, Lou K, Hua X, Huang Z, et al. Clinical value of immune-inflammatory parameters to assess the severity of coronavirus disease 2019. Int J Infect Dis. 2020 Jun;95:332-339. doi: 10.1016/j.ijid.2020.04.041.

28- RECOVERY Collaborative Group, Horby P,

Lim WS, Emberson JR, Mafham M, Bell JL, et al. Dexamethasone in Hospitalized Patients with Covid-19. N Engl J Med. 2021 Feb 25;384(8):693-704. doi: 10.1056/NEJMoa2021436.

29- Albuquerque AM, Tramujas L, Sewanan LR, Williams DR, Brophy JM. Mortality Rates Among Hospitalized Patients With COVID-19 Infection Treated With Tocilizumab and Corticosteroids: A Bayesian Reanalysis of a Previous Meta-analysis. JAMA Netw Open. 2022 Feb 1;5(2):e220548. doi: 10.1001/jamanetworkopen.2022.0548.

30- Berardicurti O, Ruscitti P, Ursini F, D'Andrea S, Ciaffi J, Meliconi R, et al. Mortality in tocilizumab-treated patients with COVID-19: a systematic review and meta-analysis. Clin Exp Rheumatol. 2020 Nov-Dec;38(6):1247-1254.

31- Gupta S, Wang W, Hayek SS, Chan L, Mathews KS, Melamed ML, et al. Association Between Early Treatment With Tocilizumab and Mortality Among Critically Ill Patients With COVID-19. JAMA Intern Med. 2021 Jan 1;181(1):41-51. doi: 10.1001/jamainternmed.2020.6252.

32- Rosas IO, Bräu N, Waters M, Go RC, Hunter BD, Bhagani S, et al. Tocilizumab in Hospitalized Patients with Severe Covid-19 Pneumonia. N Engl J Med. 2021 Apr 22;384(16):1503-1516. doi: 10.1056/NEJMoa2028700.

33- Lan SH, Lai CC, Huang HT, Chang SP, Lu LC, Hsueh PR. Tocilizumab for severe COVID-19: a systematic review and meta-analysis. Int J Antimicrob Agents. 2020 Sep;56(3):106103. doi: 10.1016/j.ijantimicag.2020.106103.

34- Tleyjeh IM, Kashour Z, Damlaj M, Riaz M, Tlayjeh H, Altannir M, et al. Efficacy and safety of tocilizumab in COVID-19 patients: a living systematic review and meta-analysis. Clin Microbiol Infect. 2021 Feb;27(2):215-227. doi: 10.1016/j.cmi.2020.10.036.

35- Bhimraj A, Morgan RL, Shumaker AH, Baden L, Cheng VCC, Edwards KM, et al. Infectious Diseases Society of America Guidelines on the Treatment and Management of Patients with COVID-19. Clin Infect Dis. 2022 Sep 5:ciac724. doi: 10.1093/cid/ciac724.

36- Available from: https://www.covid19treatmentguidelines.nih.gov/

Downloaded from on 8/30/2022

37- Lakatos B, Szabo BG, Bobek I, Gopcsa L, Beko G, Kiss-Dala N, et al. Laboratory parameters predicting mortality of adult in-patients with COVID-19 associated cytokine release syndrome treated with high-dose tocilizumab. Acta Microbiol Immunol Hung. 2021 Aug 6. doi: 10.1556/030.2021.01526.

38- Hashimoto S, Yoshizaki K, Uno K, Kitajima H, Arai T, Tamura Y, et al. Prompt Reduction in CRP, IL-6, IFN-γ, IP-10, and MCP-1 and a Relatively Low Basal Ratio of Ferritin/CRP Is Possibly Associated With the Efficacy of Tocilizumab Monotherapy in Severely to Critically Ill Patients With COVID-19. Front Med (Lausanne). 2021 Sep 23;8:734838. doi: 10.3389/fmed.2021.734838.

39- Şener MU, Çiçek T, Öztürk A. Highlights of clinical and laboratory parameters among severe COVID-19 patients treated with tocilizumab: a retrospective observational study. Sao Paulo Med J. 2022 Sep-Oct;140(5):627-635. doi: 10.1590/1516-3180.2021.0604.R1.23112021.

40- Khurshid S, Rehman N, Ahmed S, Ahmad B, Khurshid M, Muhammad A, et al. Early Fall in C-Reactive Protein (CRP) Level Predicts Response to Tocilizumab in Rapidly Progressing COVID-19: Experience in a Single-Arm Pakistani Center. Cureus. 2021 Nov 30;13(11):e20031. doi: 10.7759/cureus.20031.

41- Guz D, Gafter-Gvili A, Lev N, Sahaf Levin G, Lev S. Tocilizumab Treatment Effect on Iron Homeostasis in Severe COVID-19 Patients. Acta Haematol. 2022;145(4):440-447. doi: 10.1159/000522307.

42- Nyati A, Gokhroo A, Sharma G. Association of injection Tocilizumab and inflammatory markers in COVID19. J Assoc Physicians India. 2022 Apr;70(4):11-12.

43- Pagkratis K, Chrysikos S, Antonakis E, Pandi A, Kosti CN, Markatis E, et al. Predictors of Mortality in Tocilizumab-Treated Severe COVID-19. Vaccines (Basel). 2022 Jun 20;10(6):978. doi: 10.3390/vaccines10060978.

44- WHO Rapid Evidence Appraisal for COVID-19 Therapies (REACT) Working Group, Shankar-Hari M, Vale CL, Godolphin PJ, Fisher D, Higgins JPT, et al. Association Between Administration of IL-6 Antagonists and Mortality

Among Patients Hospitalized for COVID-19: A Meta-analysis. JAMA. 2021 Aug 10;326(6):499-518. doi: 10.1001/jama.2021.11330.

45- Sheppard M, Laskou F, Stapleton PP, Hadavi S, Dasgupta B. Tocilizumab (Actemra). Hum Vaccin Immunother. 2017 Sep 2;13(9):1972-1988. doi: 10.1080/21645515.2017.1316909.

46- Khan FA, Stewart I, Fabbri L, Moss S, Robinson K, Smyth AR, et al. Systematic review and meta-analysis of anakinra, sarilumab, siltuximab and tocilizumab for COVID-19. Thorax. 2021 Sep;76(9):907-919. doi: 10.1136/thoraxjnl-2020-215266.