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Association Of ABO And Rh Blood Groups And COVID-19 Susceptibility And Disease Severity In Turkish Population – A Retrospective Case-Control Study

Türk Popülasyonunda ABO ve Rh Kan Grupları ve COVID-19 Duyarlılığı ve Hastalık Şiddeti İlişkisi - Retrospektif Bir Vaka Kontrol Çalışması

ABSTRACT

Objective:
To determine whether there is a difference in the frequency of blood groups between the general population and COVID –19 patients and whether there is a relationship between blood groups and mortality and severity of computed tomography(CT) involvement at admission in COVID –19 patients.

Materials and Methods:
1546 COVID –19 patients were evaluated. Clinical data including age, sex, comorbidities, complaints at the time of admission, and outcome of the patients were obtained from medical records. In order to determine the normal distribution of the blood groups in the general population, a previous study conducted on Turkish population which consisted of 3,022,883 healthy blood donors of Turkish Red Crescent was identified as a control group.

Results:
There was no significant difference in gender, CT results, duration of hospitalization, and mortality rates between the blood groups (p >0.05 for all). When we compared the frequency of blood groups between the population and the patients, a statistically significant difference was found in regard to the A Rh + blood group rates (p = 0.026). Also in our study, the rate of A Rh + was found to be higher in hospitalized COVID –19 patients than the population. Also, the frequency of A Rh+ blood group was higher in patients with severe chest CT involvement compared to the normal population (p = 0.007).

Conclusion:
No significant difference was found between the blood groups and gender, severity of CT findings, duration of hospitalization, and mortality rates. But the rate of A Rh + was detected to be significantly higher in hospitalized COVID –19 patients than the population. Also, patients who had severe chest CT findings had higher frequency of A Rh+ blood group when compared to the normal population.

ÖZ
Amaç:
COVID-19 hastalarında genel popülasyon ile COVID-19 hastaları arasında kan gruplarının sıklığında bir fark olup olmadığını ve kan grupları ile mortalite ve göğüs bilgisayarlı tomografi (BT) tutulumunun şiddeti arasında bir ilişki olup olmadığını belirlemek.

Gereç ve Yöntem:
1546 COVID-19 hasta değerlendirildi. Hastaların yaş, cinsiyet, ek hastalıklar, başvuru sırasındaki şikayetleri ve sonuçları gibi klinik veriler tıbbi kayıtlardan elde edildi. Genel popülasyondaki kan gruplarının normal dağılımını belirlemek amacıyla Türk popülasyonu üzerinde yapılan ve Türk Kızılayı'nın 3.022.883 sağlıklı kan donöründen oluşan önceki bir çalışma kontrol grubu olarak belirlenmiştir.

Bulgular:
Kan grupları arasında cinsiyet, BT sonuçları, hastanede kalış süresi ve ölüm oranları açısından anlamlı fark yoktu (tümü için p>0,05). Popülasyondaki kan gruplarının sıklığı ve hastalar karşılaştırıldığında, A Rh + kan grubu oranları açısından istatistiksel olarak anlamlı bir fark bulundu (p=0,026). Çalışmamızda hastanede yatan COVID-19 hastalarında A Rh + oranı popülasyona göre daha yüksek bulunmuştur. Ayrıca, ciddi göğüs BT tutulumu olan hastalarda A Rh + kan grubu sıklığı normal popülasyona göre daha yüksekti (p=0,007).

Sonuç:
Kan grupları ile cinsiyet, BT bulgularının şiddeti, hastanede kalış süresi ve ölüm oranları arasında anlamlı bir fark bulunmadı. Ancak hastanede yatan COVID-19 hastalarında A Rh + oranının popülasyona göre önemli ölçüde daha yüksek olduğu tespit edildi. Ayrıca, ciddi göğüs BT bulguları olan hastalarda, normal popülasyona göre daha yüksek sıklıkta A Rh + kan grubu vardı.

Anahtar Sözcükler:
ABO kan grupları, Rh grupları, COVID-19, Mortalite, SARS-CoV2

INTRODUCTION
The spread of outbreak of novel coronavirus disease 2019 (coronavirus disease 2019 [COVID –19]) became an international public health emergency and World Health Organization (WHO) declared it as a pandemic on 11 March 2020. As of December 1, 2020, 62,363,527 confirmed cases of COVID –19 have been reported to the WHO, including 1,456,687 deaths (1). Several risk factors for morbidity and mortality for COVID –19 infection are determined till now, including older age, male sex, smoking and a number of chronic conditions such as diabetes mellitus, cardiovascular disease and laboratory findings (2).

ABO blood group system is found to be related to some viral infections, such as norovirus, HBV, HIV, SARS –COV-1, and MERS – COV (3-6). There are some conflicting studies suggesting that there may be a relationship between blood groups and disease severity and mortality in COVID –19 patients (7-9). While almost all of the previous studies examined the relationship between blood group and mortality and morbidity, there is no study that compares the blood groups of COVID –19 patients with the blood group rates in a large population as a control group. In this study, we aimed to determine whether there is a difference in the frequency of blood groups between the general population and COVID –19 patients and whether there is a relationship between blood groups and mortality and severity of chest CT involvement at admission in COVID –19 patients.

MATERIAL and METHODS
Study Design and Patients
This study is a retrospective cross-sectional study and consisted of 1546 RT –PCR positive COVID –19 patients who were hospitalized at Bakirkoy Dr. Sadi Konuk Training and Research Hospital in Istanbul, Turkey.

Patients were tested for SARS –CoV –2 based on epidemiological and clinical criteria as outlined in the National Guideline for the Diagnosis and Treatment Protocol for SARS CoV –2 Infection that was published and updated by Turkish Ministry of Health. Nasopharyngeal and oropharyngeal specimens were collected once from patients and specimens were tested for SARS –CoV –2 using real-time RT –PCR at our hospital. Informed consent was obtained from each subject prior to the study. The Medical Research Ethics Committee of the Bakirkoy Dr. Sadi Konuk Training and Research Hospital approved the study. We are committed to protecting patient privacy and complying with the Helsinki Declaration. (Ethical approval date:10/08/2020, Approval number: 2020-16-08). Necessary permissions were obtained from the hospital administration for the study. The medical records of 3198 patients were retrospectively analyzed. Patients who were under the age of 18, patients with unknown blood type, patients whose blood group analysis have not been done at our hospital, and patients with unknown survival status were excluded from the study. A total of 1652 patients were excluded and 1546 patients were evaluable for the final statistical analysis. Clinical data including age, sex, comorbidities, complaints at the time of admission, and outcome of the patients were obtained from medical records.

In order to determine the normal distribution of the blood groups in the general population, a previous study conducted on Turkish population which consisted of 3,022,883 healthy blood donors of Turkish Red Crescent was identified as a control group (10).

Microlate (Neo-Immucor ®) grouping method is used for ABO and Rh (D) blood groups in our hospital.

Statistical Analysis
NCSS 2007 (Number Cruncher Statistical System, Kaysville, Utah, USA) program was used for statistical analysis. Descriptive statistical methods (mean, standard deviation,

median, frequency, ratio, minimum, maximum) were used when evaluating the study data. Frequency and percentage values of categorical variables, arithmetic mean and standard deviation values of quantitative variables are presented. The conformity of quantitative data to normal distribution was examined by Shapiro-Wilk test and graphical evaluations. Kruskal Wallis test was used for comparisons of 3 or more groups that did not show normal distribution. Pearson Chi-Square test and Fisher Exact test were used for comparison of qualitative data. Type I error rate was taken as 0.05 in the study.

RESULTS

Our study was conducted on a total of 1546 cases, 46.8% (n = 723) of whom were women and 53.2% (n = 823) were men. The ages of the cases ranged from 18 to 102, with a mean of 58.19 ± 17.11 years.

Of the subjects included in the study, 19.5% (n = 301) had diabetes mellitus, 20.4% (n = 315) had hypertension, 6.6% (n = 102) had chronic obstructive pulmonary disease (COPD), 15.7% (n = 243) had coronary artery disease (CAD), 7.6% (n = 118) had cancer, 6.4% (n = 99) had chronic kidney disease, 4.3% (n = 66) had cerebrovascular disease, 1.0% (n = 15) had chronic liver disease, 1.6% (n = 24) had rheumatological disease, and 3.0% (n = 46) had other diseases.

When we examine the blood groups of the patients; 39.8% (n = 616) were A Rh+, 5.0% (n = 77) were A Rh-, 13.6% (n = 211) were B Rh+, 1.5% (n = 23) were B Rh-, 6.9% (n = 107) were AB Rh+, 0.7% (n = 10) were AB Rh-, 28.5% (n = 440) were O Rh+, 4.0% (n = 62) were O Rh-. Demographic data of the patients was shown in table I.

Table I: Distribution of Descriptive Features of the Patients.

Age (year)	Min-Max (Median)	18-102 (58)	
	Mean ± SD	58.19 ± 17.11	
Sex	Female	723	46.8
	Male	823	53.2
Comorbid disease	Diabetes Mellitus	301	19.5
	Hypertension	315	20.4
	COPD	102	6.6
	CAD	243	15.7
	Cancer	118	7.6
	CKD	99	6.4
	CVD	66	4.3
	Chronic liver disease	15	1.0
	Rheumatological disease	24	1.6
	Other diseases	46	3.0
Blood Group	A Rh+	616	39.8
	A Rh-	77	5.0
	B Rh+	211	13.6
	B Rh-	23	1.5
	AB Rh+	107	6.9
	AB Rh-	10	0.7
	O Rh+	440	28.5
	O Rh-	62	4.0

COPD: Chronic obstructive pulmonary disease,
CAD: Coronary artery disease, CKD: Chronic kidney disease,
CVD: Cerebrovascular disease,

Admission symptoms of the cases were sown in Table II; 35.7% (n = 552) had shortness of breath, cough in 28.1% (n = 434), fever in 27.5% (n = 425), weakness in 16.1% (n = 249), and loss of appetite in 11.8% (n = 182).

In regard to chest CT results, 7.0% of the cases (n = 108) had atypical findings, and typical findings are observed in 80.0% (n = 1237). When the severity of CT involvement results is examined; 17.9% (n = 222) was normal, 34.8% (n = 430) was mild, 34.9% (n = 432) was moderate and 12.4% (n = 153) were severe.

The length of stay ranges from 1 to 81 days, with an average of 10.38 ± 8.52 days and a median of 8 days. Mortality was seen in 14.6% (n = 226) of the cases. Clinical data of the patients was shown in table II.

Table II: Survival, duration of hospitalisation, and the distribution of patients' characteristics at admission.

		n	%
Admission symptoms	Fever	425	27.5
	Sore throat	150	9.7
	Cough	434	28.1
	Shortness of breath	552	35.7
	Diarrhea	126	8.2
	Headache	81	5.2
	Smell and taste loss	38	2.5
	Myalgia / arthralgia	107	6.9
	Weakness	249	16.1
	Anorexia	182	11.8
	Widespread body pain	59	3.8
	Deterioration of consciousness & other neurological symptoms	42	2.7
	Nausea and vomiting	92	6.0
	Other symptoms	8	0.5
CT Findings	Atypical findings	108	7.0
	Typical findings	1237	80.0
	No CT Scan	201	13.0
CT involvement (n=1237)	Normal	222	17.9
	Mild	430	34.8
	Moderate	432	34.9
	Severe	153	12.4
Duration of hospitalisation (Day)	Min-Max (Median)	1-81 (8)	
	Mean ± SD	10.38±8.52	
Survival	Discharged	1320	85,4
	Exitus	226	14,6

There was no significant difference in gender, CT results, duration of hospitalization, and mortality rates between blood groups (p >0.05 for all) (Table III and IV).

Table III: Association of blood groups and gender, CT findings, and mortality.

		Blood Groups								
		A Rh+	A Rh-	B Rh+	B Rh-	AB R+	AB Rh-	0 Rh+	0 Rh-	
		n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	p
Sex	Female	271 (44.0)	43 (55.8)	101 (47.9)	11 (47.8)	43 (40.2)	6 (60.0)	216 (49.1)	32 (51.6)	0.277
	Male	345 (56.0)	34 (44.2)	110 (52.1)	12 (52.2)	64 (59.8)	4 (40,0)	224 (50.9)	30 (48.4)	
CT Findings (n=1345)	Atypical	46 (8.4)	4 (6.2)	13 (7.4)	1 (4.8)	9 (9.8)	0 (0)	28 (7.3)	7 (13.2)	0.766
	Typical	500 (91.6)	61 (93.8)	162 (92.6)	20 (95.2)	83 (90.2)	8 (100)	357 (92.7)	46 (86.8)	
Typical CT Results (n=1237)	Normal	82 (16.4)	14 (23.0)	26 (16.0)	2 (10.0)	15 (18.1)	0 (0)	74 (20.7)	9 (19.6)	0.324
	Mild	174 (34.8)	22 (36.1)	53 (32.7)	6 (30.0)	25 (30.1)	5 (62.5)	128 (35.9)	17 (37)	
	Moderate	171 (34.2)	17 (27.9)	62 (38.3)	11 (55.0)	37 (44.6)	1 (12,5)	119 (33.3)	14 (30.4)	
	Severe	73 (14.6)	8 (13.1)	21 (13.0)	1 (5.0)	6 (7.2)	2 (25)	36 (10.1)	6 (13.0)	
Exitus	No	521 (84.6)	68 (88.3)	185 (87.7)	20 (87)	91 (85)	10 (100)	376 (85.5)	49 (79.0)	0.598
	Yes	95 (15.4)	9 (11.7)	26 (12.3)	3 (13)	16 (15)	0 (0)	64 (14.5)	13 (21.0)	

Pearson chi-square test

Table IV: Evaluation of Duration of Hospitalization According to Blood Groups.

| | | | Length of stay (Day) | | |
		n	Min-Max (Median)	Mean ± SD	p
	A Rh+	590	1-81 (8.5)	10.72±9.06	0.619
	A Rh-	72	1-54 (8)	10.13±7.99	
	B Rh+	204	1-60 (8)	10.58±8.74	
Blood Groups	B Rh-	21	1-28 (7)	12.43±9.08	
	AB Rh+	102	1-42 (8)	10.22±8,10	
	AB Rh-	9	3-14 (8)	8.44±3.50	
	O Rh+	420	1-52 (7)	9.65±7.56	
	O Rh-	60	1-57 (8)	11.50±10.11	

Kruskal Wallis Test

When we compared the frequency of blood groups in the population and the patients a statistically significant difference was found in regard to the A Rh + blood group rates (p = 0.026); in our study the rate of A Rh + was found to be higher in hospitalized COVID –19 patients than the population. There was no statistically significant difference in the rates of other blood types (p> 0.05 for all) (Table V).

Table V: Evaluation of the frequency of blood groups detected in our study with the frequency of blood groups in the population.

| | | Patients | | Population | | |
		n	%	n	%	p
	A Rh+	616	39.8	1121490	37.1	0.026*
	A Rh-	77	5.0	154167	5.1	0.831
	B Rh+	211	13.6	435295	14.4	0.400
Blood group	B Rh-	23	1.5	60458	2.0	0.150
	AB Rh+	107	6.9	199510	6.6	0.611
	AB Rh-	10	0.7	27206	0.9	0.292
	O Rh+	440	28.5	894773	29.6	0.326
	O Rh-	62	4.0	129984	4.3	0.575
	Total	1546	100	3022883	100	

Pearson chi-square test   *p<0.05

In our study, while there was no difference in blood group frequencies between the normal population and the patients with normal, mild, and moderate severity of chest CT involvement (p>0.05 for all), the frequency of A Rh+ blood group was higher in patients with severe chest CT involvement compared to the normal population (p = 0.007) (Table VI).

Tablo VI: Evaluation of the frequency of blood groups detected in patients with severe CT involvement with the frequency of blood groups in the population.

| | | Patients with severe CT involvement | | Population | | |
		n	%	n	%	p
	A Rh+	171	47.7	1121490	37.1	0.007**
	A Rh-	17	5.2	154167	5.1	0.942
	B Rh+	62	13.7	435295	14.4	0.812
	B Rh-	11	0.7	60458	2.0	ƒ0.381
Blood group	AB Rh+	37	3.9	199510	6.6	0.182
	AB Rh-	1	1.3	27206	0.9	ƒ0.401
	O Rh+	119	23.5	894773	29.6	0.100
	O Rh-	14	3.9	129984	4.3	0.818
	Total	432	100	3022883	100	

Pearson chi-square test   ƒFisher Exact test   **p<0.01

DISCUSSION

In our study, we did not find a significant difference in gender, CT findings, duration of hospitalization, and mortality rates between blood groups, but the rate of A Rh + was found to be significantly higher in hospitalized COVID –19 patients than the population. Also, the frequency of A Rh+ blood group was higher in patients with severe chest CT involvement compared to the normal population.

Zhao et al. (2020) showed increased susceptibility to COVID –19 infection in AB blood group than other blood groups, whereas another study conducted by Zietz and Tatonetti (2020) found the opposite result and showed a decreased COVID –19 infection rate in patients with AB blood group. In addition, Zhao et al. (2020) detected higher mortality of COVID –19 rate in A blood group, and lower mortality in O blood group, but Zeng et al. (2020) did not find such a relationship between blood type and COVID –19 mortality. The contradictory results between these studies could be due to small sample and control sizes, population heterogeneity, racial-ethnic and regional differences (11-13).

A recently published large, multi-institutional, retrospective study conducted on 1289 patients showed no relationship between ABO blood type and COVID –19 disease severity defined as intubation or death. Also, in this study, Rh + blood was associated with higher rates of testing positive for disease (14).

Another recent study conducted on 187 patients showed an increased risk for infection with SARS –CoV –2 in patients with blood group A and a decreased risk in patients with blood group O. Due to the relatively low number of patients in the study, Rh types of the patients were not examined in

that study (9). Similarly, another study investigating the genome-wide relationship of severe Covid –19 with respiratory failure, consisting of Italian and Spanish patients, found a higher risk in the A blood group and a protective effect in the O blood group compared to other blood groups (15). Another recent study, published by Arac et al. which only aimed to determine the association between the Rh blood group and Covid–19 susceptibility, was shown that the Rh (-) blood group was protective for COVID –19 infection, and the Rh (+) blood group was susceptible to COVID –19 infection (16).

In our study, contrary to the results of some previous studies, we found that the risk of infection of the O blood group with SARS CoV –2 viruses was the same with other blood groups except A Rh + patients. Similar to our study results, in a study conducted on the Swedish population, it was shown that blood types were not associated with risk of intubation or death in COVID –19 patients and there was no relationship between the O blood group and the risk of infection and mortality. In addition to these results researchers have shown that Rh+ status was associated with higher rates of testing positive. Considering our and previous study results altogether, blood type may have a less but confounded effect on infection prevalence, intubation, and death (14).

There are several molecular level hypotheses for the variable susceptibility to disease and vulnerability to severe disease. Although we did not investigate the mechanism behind the relationship, a previous study conducted by Patrice et al. reported that monoclonal or human natural anti–A antibodies might block the virus receptor interaction, thereby providing protection which could explain why individuals with blood group A were more susceptible to SARS –CoV infection (17).

CONCLUSION
As a result, we did not find a significant difference between the blood groups and gender, severity of CT findings, duration of hospitalization, and mortality rates. But the rate of A Rh + was detected to be significantly higher in hospitalized COVID –19 patients than the population. Also, patients who had severe chest CT findings had higher frequency of A Rh+ blood group when compared to the normal population.

There are some limitations in our study. Being a retrospective observational study, being performed in a single country, and not being performed blood group subgroup analysis are the leading causes of our limitations. Also, although the major ethnic group in our country (75%) consists of Turks, not studying the genetic data of the patients included in our study in order to determine the ancestry of the patients is another limitation of our study.

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Karandere F. et al.   Akd Med J 2022; 8(3)

262