Association between chronic ACE inhibitor exposure and decreased odds of severe disease in patients with COVID-19

Naci Şenkal[*,#], *Rasimcan Meral*[**,#], *Alpay Medetalibeyoğlu*[*], *Hilal Konyaoğlu*[*], *Murat Köse*[*], *Tufan Tükek*[*]

Departments of *Internal Medicine, and **General Surgery, and Medical Biology, İstanbul Faculty of Medicine, İstanbul University; İstanbul-*Turkey*

Abstract

Objective: Coronavirus disease 2019 (COVID-19) is caused by the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). Renin-angiotensin-aldosterone-system (RAAS) inhibitors may increase the expression of angiotensin-converting enzyme 2, which is the receptor for SARS-CoV-2 Spike protein. The consequences of using angiotensin-converting enzyme inhibitors (ACEi) and angiotensin receptor blockers (ARB) during the COVID-19 pandemic are unknown.

Methods: A retrospective cohort study aiming to identify the odds of severe disease (defined as either hospitalization of ≥14 days, admission to the intensive care unit, or death) associated with exposure to ACEi or ARB was conducted. Adult patients (age ≥18 years) with COVID-19 admitted to the İstanbul Faculty of Medicine Corona Center between March 9 and May 11, 2020, were included. Chronic users of ACEi, ARB, or other antihypertensive drugs were matched according to age, sex, sick days before hospitalization, comorbidities, smoking, number of antihypertensive regimens, doxazosin use, furosemide use, and serum creatinine level. Odds ratios (OR) of having severe disease were calculated.

Results: In total, 611 patients were admitted with COVID-19, confirmed by either reverse-transcriptase polymerase chain reaction or computed tomography (CT). There were 363 males, and the age ranged from 18 to 98 years, with an average age of 57±15 years. Of these, 165 participants had severe disease (53 deaths, case fatality rate: 8.7%). Among those with hypertension (n=249), ARB exposure was compatible with decreased odds (OR=0.60, 95% CI: 0.27–1.36, p=0.31) of severe disease though not statistically significant, while ACEi exposure significantly reduced the risk of severe disease (OR=0.37, 95% CI: 0.15–0.87, p=0.03). ACEi exposure was associated with milder infiltrations seen on baseline CT, lower C-reactive protein and ferritin, higher monocytes, shorter hospitalization, and less requirement for specific empirical treatments (favipiravir and meropenem).

Conclusion: Our data suggest that exposure to ACEi drugs may have favorable effects in the context of COVID-19 pneumonia. *(Anatol J Cardiol 2020; 24: 21-9)*

Keywords: coronavirus disease 2019, angiotensin-converting enzyme 2, angiotensin II receptor type 1 blockers, angiotensin-converting enzyme inhibitors

Introduction

Coronavirus disease 2019 (COVID-19) causes a potentially fatal pneumonia. It is caused by the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) and is the first known pandemic caused by a coronavirus (1-4). Attempts to contain COVID-19 to prevent an epidemic have been unsuccessful, and the number of cases is rising exponentially in many affected countries (5). An unprecedented level of international collaboration has been launched to develop vaccines and finding effective treatment. However, there has been no breakthrough in terms of treatment, and a viable vaccine may not appear before 2021 (6). In the meantime, the options to contain the virus are limited to nonpharmacological interventions and repurposed drugs.

Spike protein (S-protein) is the major virulence factor of coronaviruses and attaches to angiotensin-converting enzyme (ACE) 2 to gain entry into the cell (7, 8). ACE2 is a surface molecule found in abundance on many different cell types, including type II pneumocytes, endothelial cells, and myocardial cells (9, 10). Functionally, ACE2 is an enzyme of the renin-angiotensin-aldosterone-system (RAAS) that antagonizes the actions of its better-known homologue, the ACE (11). ACE inhibitors (ACEi) and

[#]N.Ş. and R.M. contributed equally.

Address for correspondence: Dr. Rasimcan Meral, İstanbul Üniversitesi İstanbul Tıp Fakültesi, Genel Cerrahi Anabilim Dalı, Tıbbi Biyoloji Anabilim Dalı, İstanbul-*Türkiye*
Phone: +90 537 365 30 24 E-mail: rasimcanmeral@istanbul.edu.tr

Accepted Date: 25.06.2020 **Available Online Date:** 03.07.2020

DOI:10.14744/AnatolJCardiol.2020.57431

Anatol J Cardiol 2020; 24: 21-9
DOI:10.14744/AnatolJCardiol.2020.57431

angiotensin II receptor type 1 blockers (ARB) have been shown to increase ACE2 expression (12), which led some authors to hypothesize that they increase the infectivity of SARS-CoV-2 (13). However, historical evidence shows high morbidity associated with a depletion of ACE2 in SARS and MERS infections (10, 14). Animal models have shown that acute lung injury caused by SARS-CoV can be reversed by ACEi and ARB (14, 15). Based on this, restoring ACE2 may be expected to prevent tissue damage (16-19). Early in the COVID-19 outbreak, we observed that relatively few patients in our intensive care unit (ICU) used ACEi, which prompted further investigation. We hypothesized that patients receiving ACEi as a part of their antihypertensive regimen are less likely to suffer from severe disease compared with those receiving non-RAAS inhibiting regimens.

Methods

Overview

This manuscript covers a retrospective cohort study of all adult (age ≥18) patients with COVID-19 admitted to the İstanbul Faculty of Medicine Corona Center for treatment between March 9 and May 11, 2020. The study was registered in the Ministry of Health COVID-19 research registry and approved by the İstanbul Faculty of Medicine Ethics Board.

Data collection

A standard, targeted history was obtained from all patients upon admission. The history specifically questioned travel and contact history, presence and duration of fever, coughing, shortness of breath, sputum, fatigue and myalgia, nausea, diarrhea, and anosmia. The following comorbidities were questioned in all patients: hypertension, diabetes mellitus, chronic obstructive pulmonary disease (COPD) or asthma, coronary artery disease (CAD), congestive heart failure (CHF), and hematologic or solid malignancies. In addition, the presence of chronic kidney disease (CKD) was obtained retrospectively from patient charts. All patients were tested for COVID-19 using both real-time reverse-transcriptase polymerase chain reaction (RT-PCR) and an ultra low-dose spiral computed tomography (CT) of the chest. RT-PCR was performed using nasopharyngeal swabs (20). CT scans were evaluated by a committee made of an internist, an infectious disease specialist, and a pulmonologist. They graded the CTs arbitrarily depending on the level of infiltrations (non-COVID, mild, moderate, or severe). Vital signs were obtained immediately on presentation. Height, weight, and body mass index (BMI) measurements could not be streamlined into the triage process and were only obtained retrospectively at a follow-up visit after the patients were cured from the disease. A complete blood count, full biochemistry, coagulation assay including D-dimer, inflammation markers [C-reactive protein (CRP), procalcitonin, ferritin, fibrinogen], cardiac troponin T, and pro-brain natriuretic peptide were obtained as part of routine patient care. Antihypertensive treatment history was collected from the social security institution (Sosyal Güvenlik Kurumu, SGK) digital prescription system (MEDULA).

In-patient treatment

Patients were admitted and treated according to the published protocols of the Turkish Ministry of Health (21). Involvement in this study did not influence the treatment the patients received. All treatments were off-label and empirical, as there was no effective treatment of COVID-19 at the time. A late intubation strategy was adopted. Patients younger than 50 years of age with mild symptoms were sent home for self-isolation and were beyond the scope of this study. Standard treatment included hydroxychloroquine and azithromycin. Low-molecular-weight heparin (enoxaparin or bemiparin) and dipyridamole were added to all patients with no contraindications later in the outbreak (after April 2, 2020). Other specific treatments were used depending on the clinical course of the patient. Common specific treatments included favipiravir, tocilizumab, anakinra, meropenem, and piperacillin/tazobactam.

Outcomes

Patients were divided into two groups based on their outcomes: severe and nonsevere. Severe COVID-19 infection was defined as either hospitalization of ≥14 days, admission to the ICU, or all-cause death. Nonsevere infection was defined as event-free discharge from the hospital. Discharge of patients from the corona center and admission of patients to the ICU were clinical decisions made as part of the patients' clinical care, independent of this study.

Matching

To control for confounding factors, 1:1 matching was used (22). Two separate matches were performed, yielding three groups in total. ACEi users had the smallest sample size, and thus were used as the cases. ARB and non-RAAS inhibitors users were used as controls in two separate matching runs, yielding three similar groups (ACEi, ARB, and non-RAAS inhibitors). The non-RAAS inhibitors group was used as the principal control. Patients using >3 antihypertensives at the same time, both an ACEi and ARB at the same time, or an aldosterone antagonist (spironolactone) were excluded. Cases were matched to controls according to age, sex, sick days before hospital admission, comorbidities (diabetes mellitus, COPD/asthma, CAD, CHF, and CKD), current smoking status, number of antihypertensives used, furosemide use, doxazosin use, and serum creatinine level. OR of having a severe disease were calculated.

Statistical analyses

Normal distribution was tested using Kolmogorov–Smirnov test. For parameters with nonnormal distribution, ranks were compared using Wilcoxon's rank-sum test or Kruskal–Wallis rank-sum test. For normally distributed parameters, means

were compared using either t-test or one-way analysis of variance test. Tukey's post hoc test was performed on significant variables. Frequencies were compared using Fisher's exact test and presented as frequencies and percentages. Continuous data were presented as either mean±SD or median (interquartile range) unless otherwise stated. A p-value of <0.05 was considered statistically significant. Analyses were performed using R statistical software v4.0.0 (Vienna, Austria). Odds ratios (OR) were calculated using the EpiTools package v0.5.10.1. Matching was performed using the matchControls() function of e1071 package v1.7.3, which uses the daisy pairwise dissimilarity algorithm based on Gower's coefficient (23, 24). Data and code for all analyses have been shared in a Github repository (https://github.com/meralr/COVID_ISTANBUL).

Results

Study population

Between March 9 and May 11, 2020, 619 patients were admitted to the adult corona center. Of these, 611 had COVID-19 confirmed by either RT-PCR (n=363, 59%) or CT (n=594, 97%). Among these, there were 363 males and 248 females. The age ranged from 18 to 98 years, with an average age of 57±15 years. As of May 22, 2020, 165 (27%) had severe disease and 446 were discharged event-free. Among the severe disease ones, 70 were hospitalized for >14 days, 42 were admitted into the ICU, and 53 died (case fatality rate: 8.7%, 95% CI: 6.6–11%). The baseline characteristics of the study population are shown in Table 1. The severe disease group was significantly older than the nonsevere group [55 years (IQR: 45–65) vs. 64 years (IQR: 52–75), respectively]. Still, both groups overlapped substantially, and deaths were observed in participants as young as 33 years of age (Fig. 1a). Severe disease was predicted by the level of infiltrations seen on baseline CT (p<0.001). Compared with those with a normal lung or only mild infiltrations, moderate and diffuse infiltrations were 2.7 (95% CI: 1.8–4.2) and 13.1 (95% CI: 7.4–23.2) times more likely to have severe disease, respectively. Male sex, history of hypertension, CAD, CHF, and CKD were associated with increased odds of severe disease (Fig. 1b). Diabetes mellitus, COPD/asthma, or history of solid or hematologic malignancies was compatible with increased odds of severe disease, but these did not reach statistical significance (Fig. 1b). Patient-reported shortness of breath (OR=3.2, 95% CI: 2.2–4.7) and clinical respiratory distress [respiratory rate ≥22 (OR=5.9, 95% CI: 4.0–8.7), intercostal retractions and use of accessory muscles (OR=6.7, 95% CI: 4.5–10)] were strong predictors of severe disease (Fig. 1b). No significant associations were observed between BMI and any of the outcomes. However, the data was heavily biased as the data was mostly collected on follow-up visits, where the deceased cases were missed. The association between smoking status and severe disease could not be inferred from our data. Only 34% of our patients were aware of any sick contacts they may have had.

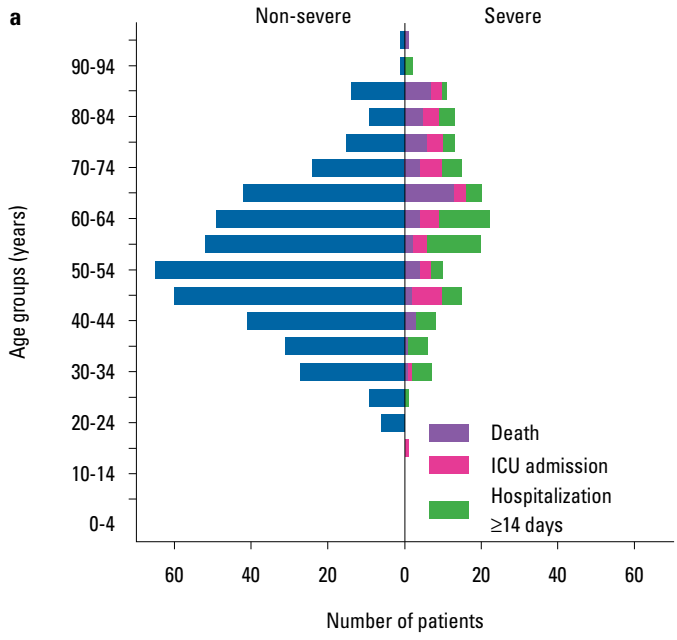
Hypertension and exposure to antihypertensives

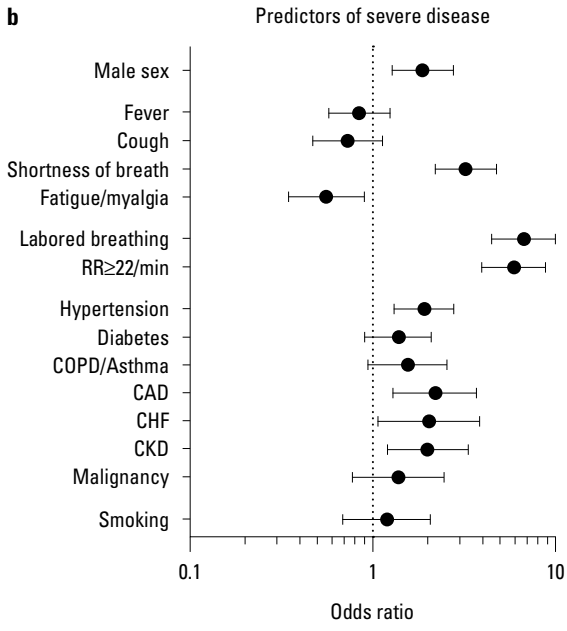
Hypertension was the most common comorbidity (n=249, 41%), 87 of whom had a severe disease course (35%; OR=1.9, 95% CI: 1.3–2.8; p<0.001). The case fatality rate of hypertensive patients was 10.8% (n=27). Those with severe disease had significantly lower baseline systolic blood pressure [median 130 (IQR: 110–143) vs. 140 (IQR: 120–145) mm Hg, respectively; p=0.024], a trend for lower diastolic blood pressure [median 75 (IQR: 70–80) vs. 80 (IQR: 70–85) mm Hg, respectively; p=0.054], and a significantly higher pulse rate [median 96 (IQR: 86–108) vs. 92 (IQR: 85–100) mm Hg, respectively; p=0.041], suggesting a state near distributive shock.

Matching was considered successful except for the number of antihypertensive regimens used by the patients (Table 2). As ACEi and ARB are commonly used in combination with thiazide diuretics, these drugs were less frequently used as single therapies, which biased the distribution. Exposure to thiazide diuretics was not associated with severe disease among the subgroup of ARB or ACEi users (n=102; crude OR=0.84, 95% CI: 0.34–2.1; p=0.82). BMI was not used as a matching parameter due to missing and biased data, but after the match, there was no significant difference observed between the groups (p=0.13). The non-RAAS inhibitors group appeared to have a lower median BMI.

The odds ratio of chronic ARB exposure was compatible with decreased risk of severe disease but did not reach statistical significance (crude OR=0.53, 95% CI: 0.29–0.98, p=0.046; adjusted OR=0.61, 95% CI: 0.27–1.4, p=0.31), while chronic ACEi exposure was found to be associated with significantly reduced risk of severe disease (crude OR=0.37, 95% CI: 0.18–0.78, p=0.012; adjusted OR=0.37, 95% CI: 0.15–0.87, p=0.034) (Fig. 1, Panels c-f). Only three patients in the ACEi group required intensive care (p=0.069), and a shorter time to discharge was observed (a median of 8 days in the ACEi group vs. 10 and 12 in the ARB and non-RAAS inhibitor groups, respectively; p=0.033). Participants chronically exposed to ACEi were 2.7 times less likely to have severe disease compared with those exposed to non-RAAS inhibitors. There was no significant difference in the RT-PCR positivity rates (p=1). Infiltrations seen on baseline CT were significantly milder in the ACEi group than in both ARB and non-RAAS inhibitors groups (p=0.041 and 0.028, respectively) (Table 2). However, ARB group did not differ from non-RAAS inhibitors group (p=0.99). The ACEi group had a trend for better blood oxygen saturation measured by a pulse oximeter on admission (p=0.074) and a slower pulse (p=0.007), with the clinical relevance of the latter remaining uncertain. Compared with both non-RAAS inhibitors and ARB groups, ACEi group had a trend to have higher lymphocytes (p=0.032 and 0.045, respectively) and significantly lower CRP (p=0.017 and p=0.004, respectively) and ferritin (p=0.014 and p=0.028, respectively) levels at baseline. We observed trends for more favorable coagulation parameters (lower D-dimer and activated partial thromboplastin time) in the ACEi group (Table 2). Patients in the ACEi group were less likely to require treatment

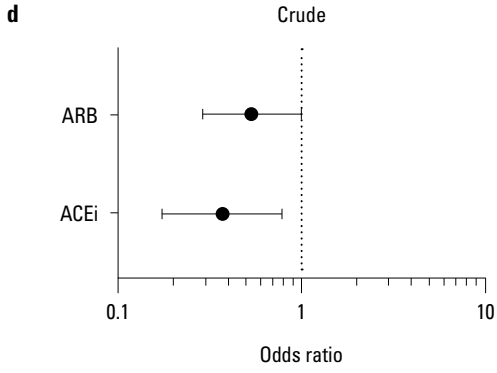
Anatol J Cardiol 2020; 24: 21-9
DOI:10.14744/AnatolJCardiol.2020.57431





c

	Not severe	Severe	Total
Non-RAAS inhibitor	42	36	78
ARB	72	33	105
ACEi	44	14	58
ARB and ACEi	1	1	2
Spironolactone	2	3	5
Total	161	87	248



e

	Not severe	Severe	Total
Non-RAAS inhibitor	30	22	52
ARB	36	16	52
ACEi	41	11	52
Total	107	49	156

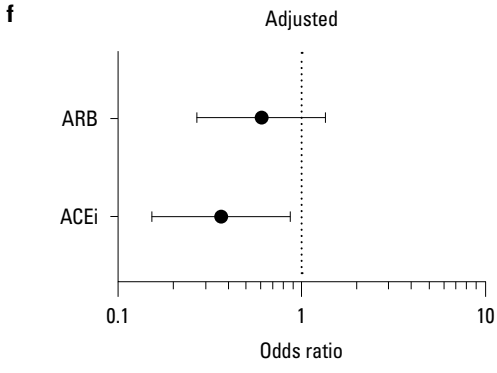


Figure 1. (a) Age distribution of patients. (b) Odds ratios of presenting features and severe disease. (c) Distribution and (d) odds ratios of severe disease associated with chronic exposure to different antihypertensive categories and in patients with a history of hypertension. (e) Distribution and (f) odds ratios of severe disease associated with chronic exposure to either ACEi or ARB in populations matched according to age, sex, comorbidities (diabetes mellitus, COPD/asthma, CAD and CHF), smoking status, and serum creatinine levels

ACEi - angiotensin-converting enzyme inhibitors; ARB - angiotensin II receptor type 1 blockers; CAD - coronary artery disease; CHF - congestive heart failure; CKD - chronic kidney disease; COPD - chronic obstructive pulmonary disease; ICU - intensive care unit

Table 1. Characteristics of coronavirus disease 2019 (COVID-19) patients recorded on admission

	Nonsevere outcome	Severe outcome	*P*-value
n	446	165	
RT-PCR positivity	260 (58%)	103 (62%)	0.40[f]
Infiltrations seen on presentation CT			
1- Normal	3	0	
2- Very mild	55	7	
3- Mild	257	58	**<0.001[w]**
4- Moderate	94	42	
5- Diffuse	26	55	
Age (years)	55±15	63±16	**<0.001[T]**
Gender (Female:Male)	198 F:248 M	50 F:115 M	**0.002[f]**
*BMI (Kg/m^2) [n missing]	28.1 (25.9-31.2) [56]	27.9 (25.5-31.1) [72]	0.45[w]
Travel history (n)	3 (1%)	3 (2%)	0.35[f]
Contact history (n)	160 (36%)	45 (27%)	**0.043[f]**
Reported symptoms			
Cough (n)	356 (81%)	124 (76%)	0.17[f]
Fever (n)	306 (70%)	108 (66%)	0.43[f]
Sputum (n)	14 (3%)	2 (1%)	0.26[f]
Shortness of breath (n)	159 (36%)	106 (65%)	**<0.001[f]**
Fatigue and/or myalgia (n)	383 (87%)	130 (79%)	**0.020[f]**
Nausea and/or vomiting (n)	74 (17%)	22 (15%)	0.61[f]
Diarrhea (n)	52 (12%)	17 (12%)	1[f]
Anosmia (n)	36 (8%)	7 (5%)	0.20[f]
Sick days before hospitalization	5 (3-7)	5 (3-7)	0.80[w]
Comorbidities and risk factors			
Hypertension (n)	162 (37%)	87 (53%)	**<0.001[f]**
# of anti-HT medications			
1	44	30	
2	65	27	
3	39	16	0.10[f]
4	12	11	
5	1	3	
Diabetes mellitus (n)	94 (21%)	45 (27%)	0.12[f]
COPD/Asthma (n)	55 (13%)	30 (18%)	0.087[f]
Coronary artery disease (n)	39 (9%)	29 (18%)	**0.004[f]**
Congestive heart failure (n)	24 (5%)	18 (11%)	**0.029[f]**
Chronic kidney disease (n)	46 (10%)	31 (19%)	**0.009[f]**
Solid malignancy (n)	28 (6%)	14 (9%)	0.37[f]
Hematologic malignancy (n)	12 (3%)	6 (4%)	0.59[f]
Smoking (n)	48 (11%)	21 (13%)	0.56[f]

*: BMI was not available at baseline and was collected at a 1-month follow-up visit. [f]: Fisher's exact test, [T]: Two samples t-test, [w]: Wilcoxon rank-sum test.
Categorical data is presented as frequency (percentages). Continuous data is presented as either mean±SD or median (Interquartile range) depending on normality.
COPD - chronic obstructive pulmonary disease; CKD - chronic kidney disease; CT - computed tomography; HT - hypertension

Anatol J Cardiol 2020; 24: 21-9
DOI:10.14744/AnatolJCardiol.2020.57431

Table 2. Characteristics of matched groups with exposure to different antihypertensive classes

	Non-RAAS inhibitors	ARB	ACEi	*P*-value
Matching parameters				
n	52	52	52	
Age (years)	65±12	63±11	63±13	0.64[A]
Gender (Female:Male)	22F:30M	26F:26M	25F:27M	0.78[f]
Sick days before hospitalization	3 (2-5)	5 (2-7)	3 (2-5)	0.51[k]
Diabetes mellitus history (n)	20 (38%)	22 (42%)	22 (42%)	0.94[f]
COPD/asthma history (n)	9 (17%)	7 (13%)	6 (12%)	1[f]
Coronary Artery Disease history (n)	14 (27%)	11 (21%)	16 (31%)	0.57[f]
Congestive heart failure (n)	6 (12%)	3 (6%)	5 (10%)	0.69[f]
Chronic kidney disease	5 (10%)	6 (12%)	4 (8%)	0.75[f]
Smoking status (n)	1 (2%)	3 (6%)	3 (6%)	0.70[f]
# of anti-HT medications (n)				
1	31[1,2]	10[1]	9[2]	
2	18	25	27	**0.007[f]**
3	3	17	16	
Furosemide use	1 (2%)	1 (2%)	2 (4%)	0.70[f]
Doxazosin use	2 (4%)	0 (0%)	1 (2%)	0.77[f]
Serum Creatinine (mg/dL)	0.9 (0.7-1.3)	0.9 (0.7-1.2)	0.9 (0.7-1.1)	1[k]
Antihypertensive medication				
Calcium channel blockers (n)	30 (58%)[2]	21 (40%)	16 (31%)[2]	**0.020[f]**
Beta blockers (n)	32 (62%)[1,2]	10 (19%)1	19 (37%)[2]	**<0.001[f]**
Thiazide diuretics (n)	3 (6%)[1,2]	26 (50%)1	19 (37%)[2]	**<0.001[f]**
ARB or ACEi type (Nonsevere:Severe)	-	Valsartan 13:7 Kandesartan 6:5 Olmesartan 6:1 Others 11:3	Ramipril 21:7 Perindopril 13:1 Others 7:3	
Characteristics				
*BMI (Kg/m^2) [n missing]	27.0 (25.7-30.5) [14]	29.5 (26.3-32) [14]	29.3 (26.8-32.5) [7]	0.13[k]
RT-PCR positivity (n)	30 (58%)	30 (58%)	31 (60%)	1[f]
Infiltrations on CT scan (n)				
1. Normal	0	0	1	
2. Very mild	5	10	11	
3. Mild	27	18	28	0.052[k]
4. Moderate	9	14	8	
5. Diffuse	10	10	4	
Systolic blood pressure (mm Hg)	138 (120-150)	140 (120-146)	140 (130-145)	0.81[k]
Diastolic blood pressure (mm Hg)	80 (70-85)	78 (70-90)	78 (70-86)	0.98[k]
Pulse rate (per minute)	92 (88-102)[2]	98 (88-103)[3]	88 (84-95)[2,3]	**0.007[k]**
Respiratory rate (per minute)	20 (16-24)	20 (17-24)	18 (16-20)	0.14[k]
SpO_2 on room air (%)	95 (92-97)	95 (90-97)	96 (94-98)	0.074[k]
**Labored breathing (n)	21 (40%)	23 (44%)	13 (25%)	0.097[f]
White blood cells (cells/µl)	6610 (4328-8485)	6980 (5078-9815)	7040 (5838-10495)	0.12[k]

Table 2. Cont.

	Non-RAAS inhibitors	ARB	ACEi	*P*-value
Neutrophils (cells/µl)	4550 (3088-6370)	5190 (3312-7758)	5235 (3492-8180)	0.28[k]
Lymphocytes (cells/µl)	915 (625-1390)	965 (650-1530)	1205 (905-1602)	0.054[k]
Monocytes (cells/µl)	430 (310-625)[2]	460 (308-725)	600 (460-762)[2]	**0.017[k]**
Aspartate transaminase (U/L)	26 (19-36)	25 (18-40)	24 (17-39)	0.92[k]
Alanine transaminase (U/L)	22 (15-30)	20 (14-31)	22 (15-38)	0.70[k]
Pro-BNP (pg/mL)	284 (101-1725)	190 (65-710)	188 (55-1309)	0.46[k]
Troponin T (pg/mL)	15 (5-25.2)	9 (4-18)	10.7 (6-31.2)	0.26[k]
C-reactive protein (mg/L)	52 (17-157)[2]	64 (32-133)[3]	28 (12-66)[2,3]	**0.009[k]**
Procalcitonin (ng/mL)	0.12 (0.05-0.38)	0.11 (0.05-0.27)	0.08 (0.05-0.17)	0.37[k]
INR	1.0 (0.9-1.0)	1.0 (0.9-1.0)	1.0 (0.9-1.0)	0.67[k]
aPTT (seconds)	29 (27-33)	29 (26-31)	28 (26-30)	0.089[k]
Ferritin (ng/mL)	334 (170-1005)[2]	334 (144-751)[3]	188 (109-366)[2,3]	**0.025[k]**
D-dimer (µg/L)	1040 (618-1658)	985 (595-1882)	665 (438-1238)	0.069[k]
Specific treatments received				
Hydroxychloroquine (n)	52 (100%)	52 (100%)	52 (100%)	1[f]
Azithromycin (n)	52 (100%)	52 (100%)	52 (100%)	1[f]
Favipiravir (n)	22 (46%)[2]	22 (45%)[3]	9 (18%)[2,3]	**<0.001[f]**
Tocilizumab (n)	13 (25%)	14 (27%)	6 (12%)	0.12[f]
Anakinra (n)	8 (17%)	3 (6%)	3 (6%)	0.15[f]
Meropenem (n)	10 (21%)	14 (29%)[3]	4 (8%)[3]	**0.026[f]**
Piperacillin/Tazobactam (n)	5 (10%)	3 (6%)	2 (4%)	0.43[f]
Outcomes				
Severe disease	22 (42%)	16 (31%)	11 (21%)	0.081[f]
Death (n)	5 (10%)	5 (10%)	2 (4%)	0.49[f]
Intensive care (n)	11 (21%)	9 (17%)	3 (6%)	0.069[f]
Hospitalization ≥14 days	22 (42%)[2]	16 (31%)	10 (19%)[2]	**0.042[f]**
Days hospitalized	12 (7-19)[2]	10 (5-16)	8 (5-12)[2]	**0.033[k]**

*: BMI was not available at baseline and was collected at a 1-month follow-up visit.
**: Labored breathing was defined as the presence of intercostal retractions and/or the use of accessory muscles as seen on inspection.
[1]: The difference between non-RAAS inhibitors and ARB is statistically significant.
[2]: The difference between non-RAAS inhibitors and ACEi is statistically significant.
[3]: The difference between ARB and ACEi is significant.
[A]: One-way Analysis of Variance [f]: Fisher's exact test [k]: Kruskal-Wallis rank-sum test.
Categorical data is presented as frequency (percentages). Continuous data is presented as either mean ± SD or median (Interquartile range) depending on normality.
ACEi - angiotensin-converting enzyme inhibitor; aPTT- activated partial thromboplastin time; ARB - angiotensin II receptor type 1 blockers; BNP - brain natriuretic peptide; COPD - chronic obstructive pulmonary disease; CKD- chronic kidney disease; CT - computed tomography; HT- hypertension; INR - international normalized ratio; RAAS - renin-angiotensin-aldosterone-system; RT-PCR - reverse-transcriptase polymerase chain reaction

with favipiravir (p<0.001) and meropenem (p=0.026), which were empirical second line agents only administered to patients with a more severe clinical course.

Discussion

In the context of COVID-19 disease, our study suggests that chronic exposure to ACE inhibitors or ARB is compatible with decreased risk of adverse outcomes such as death, ICU admission, or lengthy hospitalization. Chronic ACEi exposure was significantly associated with decreased odds of severe disease. This finding has been supported by clinical parameters known to be associated with favorable prognosis, such as milder infiltrations seen on CT scan, better saturation on admission, lower acute phase reactants (CRP and ferritin), and a more favorable coagulation profile.

Early during the COVID-19 pandemic, some authors hypothesized that ACEi and ARB enhanced the infectivity of SARS-CoV-2 due to the induction of ACE2 expression (13, 25). This

Anatol J Cardiol 2020; 24: 21-9
DOI:10.14744/AnatolJCardiol.2020.57431

hypothesis has been picked up by the media and amplified through social media, causing many patients with hypertension to discontinue their treatment out of fear. The subject has been a matter of debate, and several authors argued strongly that based on mechanistic evidence, these drugs might actually be protective rather than harmful (17-19, 26, 27). Angiotensin II has proinflammatory, profibrotic, vasoconstrictor, and prothrombotic effects through the Angiotensin type 1 (AT1) receptor, all of which are mechanisms that potentially explain complications associated with severe COVID-19 infection (26, 27). It therefore makes sense that the reduction of angiotensin II levels or the inhibition of the AT1 receptor would not only reduce these harmful effects but also enhance potentially favorable effects associated with the shunting of the RAAS pathway to increase the levels of Angiotensin 1-9 and Angiotensin 1-7, which promote anti-inflammation, antifibrosis, vasodilation, and antithrombosis through the MAS receptor (27). In contrast to ARB, ACEi may be causing further accumulation of Angiotensin 1-7, as ACE also plays a role in the breakdown of the latter (27). However, evidence for the efficacy of ARB or ACEi has been lacking except in animal models and observational studies on SARS-CoV (10, 14, 15). Large retrospective analyses found no evidence of increased or decreased COVID-19 risk associated with ACEi or ARB use (28, 29). Mehra et al. (30) found a reduced risk of death related to ACEi exposure, but the authenticity of this study was questioned, and their papers have been retracted (31). The authors' claim to have included 346 patients with PCR-confirmed COVID-19 hospitalized before March 15, 2020 from Turkey seems unlikely, as the first wave of infections had not yet hit the country (32). To our knowledge, our study is the first to demonstrate potentially favorable effects associated with ACEi after the discrediting of Mehra et al. (30).

Although our study suggests favorable outcomes associated with chronic ACEi exposure, several limitations need to be taken into consideration. First, due to the retrospective nature of this study, it is not possible to rule out all confounders. BMI may have been a critical covariate to account for, but the data was incomplete. Secondly, 41% of our patients lacked PCR or serological confirmation of SARS-CoV-2 infection, which was similar to numbers reported around the globe and was probably related to the collection of the inadequate material (33). The possibility of misdiagnoses was slim but could not be ruled out. We found that low dose chest CT was highly sensitive to detect typical COVID-19 pneumonia (we assumed high positive predictive value in the setting of a pandemic), which also gave a good idea on the prognosis, allowing aggressive treatment to be initiated immediately. Thirdly, the use of any given medication may be associated with potential confounders that were not accounted for, such as the proneness to having a dry cough associated with ACE inhibition and bradykinin accumulation. Finally, in our setting, the use of ACEi or ARB may casually be an indicator of simply receiving better medical care. Initiation of these drugs requires careful monitoring of serum creatinine levels on follow-up visits and is typically avoided in patients with poor adherence to medical advice. Drug compliance rates of patients could not be ascertained, and the adverse outcomes observed in the non-RAAS inhibitor group may be associated with uncontrolled hypertension or poor overall health status rather than the choice of drug. However, although the subgroups were too small for any meaningful comparisons, perindopril appeared as the most beneficial ACEi in our data. Perindopril is known to have a pharmacodynamic effects that persists beyond 24 hours, and studies have shown that treatment with perindopril is quite robust to missed dose effects (34).

Conclusion

In conclusion, we found no harm associated with ACEi or ARB use. Potential beneficial effects were observed in the group chronically exposed to ACEi but this remains to be proven by randomized clinical trials, as confounders could not be ruled out. Regardless, our data support the recommendation to continue antihypertensive regimens uninterrupted.

Acknowledgements: N.Ş. and R.M. contributed equally. T.T. and M.K. accept full responsibility for the accuracy and integrity of the data as the guarantors of this work. All authors edited and approved the final version of the manuscript.

No funding has been received for this study. R.M. was supported by the Scientific and Technical Research Council of Turkey (TUBITAK-BIDEB: 2211-A) and Turkish Council of Higher Education (YÖK 100/2000). Patient care, tests, and the infrastructural changes associated with the pandemic were funded by the Turkish Ministry of Health. All authors have completed the Unified Competing Interest form (available on request from the corresponding author) and declare no competing interests directly related to this work. Outside the submitted work, N.Ş. reports travel grants from Novartis and Sandoz; A.M. reports travel grants from Sanofi and Sanovel; M.K. reports travel grants and honoraria from Astra Zeneca, Boehringer Ingelheim, and İbrahim Etem Menarini; T.T. reports honoraria for presentations from Astra Zeneca, Boehringer Ingelheim, Sanofi, Novartis, Novo Nordisk and Merck.

We are grateful to Prof. Dr. Cemil Tasçıoğlu and Prof. Dr. Seyit Mehmet Kayacan, who dedicated their lives (and death) to teaching thousands of medical students and residents among all other healthcare workers whom we lost to COVID-19. We thank Dr. Timur Selçuk Akpınar for his valuable input and help with editing the language.

Conflict of interest: None declared.

Peer-review: Externally peer-reviewed.

Authorship contributions: Concept – N.Ş., R.M., A.M., M.K., T.T.; Design – R.M.; Supervision – M.K., T.T.; Fundings – None; Materials – None; Data collection and/or processing – N.Ş., A.M., H.K.; Analysis and/or interpretation – R.M.; Literature search – N.Ş., R.M., A.M., M.K.; Writing – N.Ş., R.M., A.M.; Critical review – N.Ş., R.M., A.M., H.K., M.K., T.T.

References

1. Zhou P, Yang XL, Wang XG, Hu B, Zhang L, Zhang W, et al. A pneumonia outbreak associated with a new coronavirus of probable bat origin. Nature 2020; 579: 270-3. [CrossRef]
2. Wu F, Zhao S, Yu B, Chen YM, Wang W, Song ZG, et al. A new coronavirus associated with human respiratory disease in China. Nature 2020; 579: 265-9. [CrossRef]
3. Li Q, Guan X, Wu P, Wang X, Zhou L, Tong Y, et al. Early Transmission Dynamics in Wuhan, China, of Novel Coronavirus-Infected Pneumonia. N Engl J Med 2020; 382: 1199-207. [CrossRef]
4. WHO. WHO Director-General's opening remarks at the media briefing on COVID-19 2020. Available from: URL; https://www.who.int/dg/speeches/detail/who-director-general-s-opening-remarks-at-the-media-briefing-on-covid-19---11-march-2020
5. The Center for Systems Science and Engineering (CSSE) at Johns Hopkins University. Coronavirus COVID-19 Global Cases 2020. Available from: URL; https://coronavirus.jhu.edu/map.html
6. Thanh Le T, Andreadakis Z, Kumar A, Gómez Román R, Tollefsen S, Saville M, et al. The COVID-19 vaccine development landscape. Nat Rev Drug Discov 2020; 19: 305-6. [CrossRef]
7. Li W, Moore MJ, Vasilieva N, Sui J, Wong SK, Berne MA, et al. Angiotensin-converting enzyme 2 is a functional receptor for the SARS coronavirus. Nature 2003; 426: 450-4. [CrossRef]
8. Wrapp D, Wang N, Corbett KS, Goldsmith JA, Hsieh CL, Abiona O, et al. Cryo-EM structure of the 2019-nCoV spike in the prefusion conformation. Science 2020; 367: 1260-3. [CrossRef]
9. Hamming I, Timens W, Bulthuis ML, Lely AT, Navis G, van Goor H. Tissue distribution of ACE2 protein, the functional receptor for SARS coronavirus. A first step in understanding SARS pathogenesis. J Pathol 2004; 203: 631-7. [CrossRef]
10. Oudit GY, Kassiri Z, Jiang C, Liu PP, Poutanen SM, Penninger JM, et al. SARS-coronavirus modulation of myocardial ACE2 expression and inflammation in patients with SARS. Eur J Clin Invest 2009; 39: 618-25. [CrossRef]
11. Nicholls J, Peiris M. Good ACE, bad ACE do battle in lung injury, SARS. Nat Med 2005; 11: 821-2. [CrossRef]
12. Ferrario CM, Jessup J, Chappell MC, Averill DB, Brosnihan KB, Tallant EA, et al. Effect of angiotensin-converting enzyme inhibition and angiotensin II receptor blockers on cardiac angiotensin-converting enzyme 2. Circulation 2005; 111: 2605-10. [CrossRef]
13. Sommerstein R. Rapid response: re: preventing a covid-19 pandemic: ACE inhibitors as a potential risk factor for fatal Covid-19. BMJ 2020; 368: m810. [CrossRef]
14. Kuba K, Imai Y, Rao S, Gao H, Guo F, Guan B, et al. A crucial role of angiotensin converting enzyme 2 (ACE2) in SARS coronavirus-induced lung injury. Nat Med 2005; 11: 875-9. [CrossRef]
15. Imai Y, Kuba K, Rao S, Huan Y, Guo F, Guan B, et al. Angiotensin-converting enzyme 2 protects from severe acute lung failure. Nature 2005; 436: 112-6. [CrossRef]
16. Zhang H, Penninger JM, Li Y, Zhong N, Slutsky AS. Angiotensin-converting enzyme 2 (ACE2) as a SARS-CoV-2 receptor: molecular mechanisms and potential therapeutic target. Version 2. Intensive Care Med 2020; 46: 586-90. [CrossRef]
17. Gurwitz D. Angiotensin receptor blockers as tentative SARS-CoV-2 therapeutics. Drug Dev Res 2020: 10.1002/ddr.21656. doi: 10.1002/ddr.21656. Epub ahead of print [CrossRef]
18. Kuster GM, Pfister O, Burkard T, Zhou Q, Twerenbold R, Haaf P, et al. SARS-CoV2: should inhibitors of the renin-angiotensin system be withdrawn in patients with COVID-19? Eur Heart J 2020; 41: 1801-3.
19. Tignanelli CJ, Ingraham NE, Sparks MA, Reilkoff R, Bezdicek T, Benson B, et al. Antihypertensive drugs and risk of COVID-19? Lancet Respir Med 2020; 8: e30-1. [CrossRef]
20. Marty FM, Chen K, Verrill KA. How to Obtain a Nasopharyngeal Swab Specimen. N Engl J Med 2020; 382: e76. [CrossRef]
21. COVID-19 Erişkin Tedavi Algoritması: Türkiye Cumhuriyeti Saglık Bakanlığı; 2020. Available from: https://covid19bilgi.saglik.gov.tr/depo/algoritmalar/COVID19-PLKACILHASTAYONETIMI.pdf
22. Stuart EA. Matching methods for causal inference: A review and a look forward. Stat Sci 2010; 25: 1-21. [CrossRef]
23. Gower JC. A general coefficient of similarity and some of its properties. Biometrics 1971; 27: 857-71. [CrossRef]
24. Kaufman L, Rousseeuw PJ. Finding groups in data: an introduction to cluster analysis: John Wiley & Sons; 2009.
25. Watkins J. Preventing a covid-19 pandemic. BMJ 2020; 368: m810.
26. Vaduganathan M, Vardeny O, Michel T, McMurray JJV, Pfeffer MA, Solomon SD. Renin-Angiotensin-Aldosterone System Inhibitors in Patients with Covid-19. N Engl J Med 2020; 382: 1653-9. [CrossRef]
27. Lubel J, Garg M. Renin-Angiotensin-Aldosterone System Inhibitors in Covid-19. N Engl J Med 2020; 382: e92. [CrossRef]
28. Mancia G, Rea F, Ludergnani M, Apolone G, Corrao G. Renin-Angiotensin-Aldosterone System Blockers and the Risk of Covid-19. N Engl J Med 2020; 382: 2431-40. [CrossRef]
29. Reynolds HR, Adhikari S, Pulgarin C, Troxel AB, Iturrate E, Johnson SB, et al. Renin-Angiotensin-Aldosterone System Inhibitors and Risk of Covid-19. N Engl J Med 2020; 382: 2441-8. [CrossRef]
30. Mehra MR, Desai SS, Kuy S, Henry TD, Patel AN. Cardiovascular Disease, Drug Therapy, and Mortality in Covid-19. N Engl J Med 2020; 382: e102. [CrossRef]
31. Rubin EJ. Expression of Concern: Mehra MR et al. Cardiovascular Disease, Drug Therapy, and Mortality in Covid-19. N Engl J Med. DOI: 10.1056/NEJMoa2007621. N Engl J Med 2020; 382: 2464. [CrossRef]
32. Watson JA, Meral R, Price R, Simpson J. An open letter to Mehra et al and The New England Journal of Medicine (Version 1). Zenodo. http://doi.org/10.5281/zenodo.3873178
33. Sethuraman N, Jeremiah SS, Ryo A. Interpreting Diagnostic Tests for SARS-CoV-2. JAMA 2020; doi: 10.1001/jama.2020.8259. Epub ahead of print. [CrossRef]
34. Tan KW, Leenen FH. Persistence of anti-hypertensive effect after missed dose of perindopril. Br J Clin Pharmacol 1999; 48: 628-30.