Impact of ACE2 overexpressing drugs in covid-19 severity. A retrospective study

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ABSTRACT

Objective and location: Angiotensin II receptor blockers (ARBs), angiotensin-converting enzyme inhibitors (ACEi) and non-steroidal anti-inflammatory drugs (NSAIDs) may aggravate the evolution of patients with COVID-19 because they would up-regulate angiotensin-converting enzyme receptors (ACE2) in target cells, thereby facilitating the SARS-CoV-2 infection. In order to corroborate this hypothesis, the objective of this study was to determine whether patients with confirmed COVID-19 infection treated with drugs that increase ACE2 expression had a higher risk of poor disease progression than patients not treated with these drugs.

Methodology: Retrospective cohort study that included patients with a PCR-confirmed diagnosis of SARS-CoV-2 infection in the province of Alava (Spain) between February and May 2020.The sample of patients was separated into two groups:treated and NO treated with drugs that overexpress ACE2. The primary endpoint assessed was the poor COVID-19 disease progression, defined as any of the following events: pneumonia, acute pulmonary oedema, stroke, acute respiratory syndrome and hypoxaemia, cardiac involvement or thrombo-embolic phenomena.

Results: 3,060 adult patients were included. After statistical analysis, it was observed that neither ACEi nor ARBs increase significantly the risk of poor evolution disease (ACEi OR 1.245; 95% CI 0.927-1.673; p=0.146 and ARBs OR 1.345; 95% CI 0.980-1.847; p=0.067) but on the contrary; NSAIDs increase the risk of poor evolution (OR 1.466; 95% CI 1.149-1.870; p=0.002).

Conclusions: In view of the results obtained, patients treated with NSAIDs could present a higher risk of a poor evolution of COVID-19. It would be advisable to closely monitor patients with COVID-19 who are being treated with this type of drug for the treatment of chronic pain.

Keywords: **COVID-19; SARS-CoV-2; Angiotensin-Converting Enzyme Inhibitors; non-steroidal anti-inflammatory drugs; Angiotensin Receptor Antagonists.**

RESUMEN

Objetivo: Los antagonistas de los receptores de angiotensina II (ARA II), los inhibidores de la enzima convertidora de angiotensina (IECA) y los antiinflamatorios no esteroideos (AINE) pueden agravar la evolución de los pacientes con COVID-19 porque regularían positivamente los receptores de la enzima convertidora de angiotensina (ACE2) en las células diana, facilitando así la infección por SARS-CoV-2. Para confirmar esta hipótesis, el objetivo de este estudio fue determinar si los pacientes con infección confirmada por COVID-19 tratados con fármacos que aumentan la expresión de ACE2 tenían un mayor riesgo de mala evolución de la enfermedad que los pacientes no tratados con este tipo de fármacos.

Metodología: Estudio de cohorte retrospectivo en el que se incluyeron pacientes con diagnóstico confirmado mediante PCR de infección por SARS-CoV-2 en la provincia de Álava (España) entre febrero y mayo de 2020. La muestra de pacientes se dividió en dos grupos: tratados y no tratados con fármacos que sobreexpresan la ECA2. La variable principal evaluada fue la mala evolución de la enfermedad COVID-19, definida como cualquiera de los siguientes eventos: neumonía, edema pulmonar agudo, ictus, insuficiencia respiratoria aguda, síndrome de dificultad respiratoria e hipoxemia, afectación cardíaca o fenómenos tromboembólicos.

Resultados: Se incluyeron 3.060 pacientes. Tras el análisis estadístico, se observó que ni los IECA ni los ARA II aumentan significativamente el riesgo de mala evolución de la enfermedad (IECA OR 1,245; IC 95% 0,927-1,673; p=0,146 y ARAII OR 1,345; IC 95% 0,980-1,847; p=0,067), por el contrario, los AINE aumentan el riesgo de mala evolución (OR 1,466; IC 95% 1,149-1,870; p=0,002).

Conclusiones: Los pacientes tratados con AINE podrían presentar mayor riesgo de mala evolución de la COVID-19. Sería recomendable realizar un estrecho seguimiento de los pacientes con COVID-19 que estén siendo tratados estos fármacos para el tratamiento del dolor crónico.

Palabras clave: **COVID-19; SARS-CoV-2; inhibidores de la enzima convertidora de angiotensina; fármacos anti-inflamatorios no esteroideos; antagonistas de los receptores de angiotensina.**

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INTRODUCTION

The SARS-CoV-2 pandemic has originated an unprecedented strain on healthcare systems across de globe. On 9 February 2024, the number of diagnosed cases worldwide was 702879693 accounting for 6980517 deaths[1]. The most common symptoms are fever, cough, shortness of breath, and fatigue. Other symptoms such as aches, nasal congestion, rhinorrhoea, sore throat or diarrhoea[2], hypogeusia (5.6%) and hyposmia (5.1%) have also been reported[3].These symptoms are usually mild and appear gradually, however one among six patients can develop severe disease symptoms[2].

Several risk factors for severe disease have been identified as male sex, age over 60-65 years, comorbidities such as hypertension, diabetes, cardiovascular and cerebrovascular disease and chronic obstructive pulmonary disease (COPD)[3-5]. Importantly, many of these comorbidities are often treated with angiotensin-converting enzyme (ACE) inhibitors[6]. Smoking and obesity have also been associated with an increased risk of severe disease[7,8].

Human pathogenic coronaviruses bind to their target cells via angiotensin II-converting enzyme (ACE2)[9]. Concerns about whether angiotensin II receptor blockers (ARBs) and angiotensin-converting enzyme inhibitors (ACEi) may have deleterious effects on morbidity and mortality in patients with COVID-19 are based on the hypothesis that these drugs could upregulate ACE2 in target cells, thereby facilitating SARS-CoV-2 infection[10]. Moreover, ibuprofen and all non-steroidal anti-inflammatory drugs (NSAIDs) in general could also promote ACE2 receptor expression on lung cells[11].

In recent years, several studies have been published on the potential cardiovascular, pulmonary and renal benefits of ACE2 that have been linked to its suppressive effect on Angiotensin II and its conversion to Angiotensin, which has potentially beneficial vasodilator and anti-inflammatory properties in general, and especially in the context of the COVID-19 pandemic[9,12]. According to these studies the SARS-CoV-2 infection could alter ACE2 expression in cells, thereby altering the physiological balance between ACE/ACE2 and angiotensin II/angiotensin I[9,12] and subsequently causing severe organ damage[9,12].

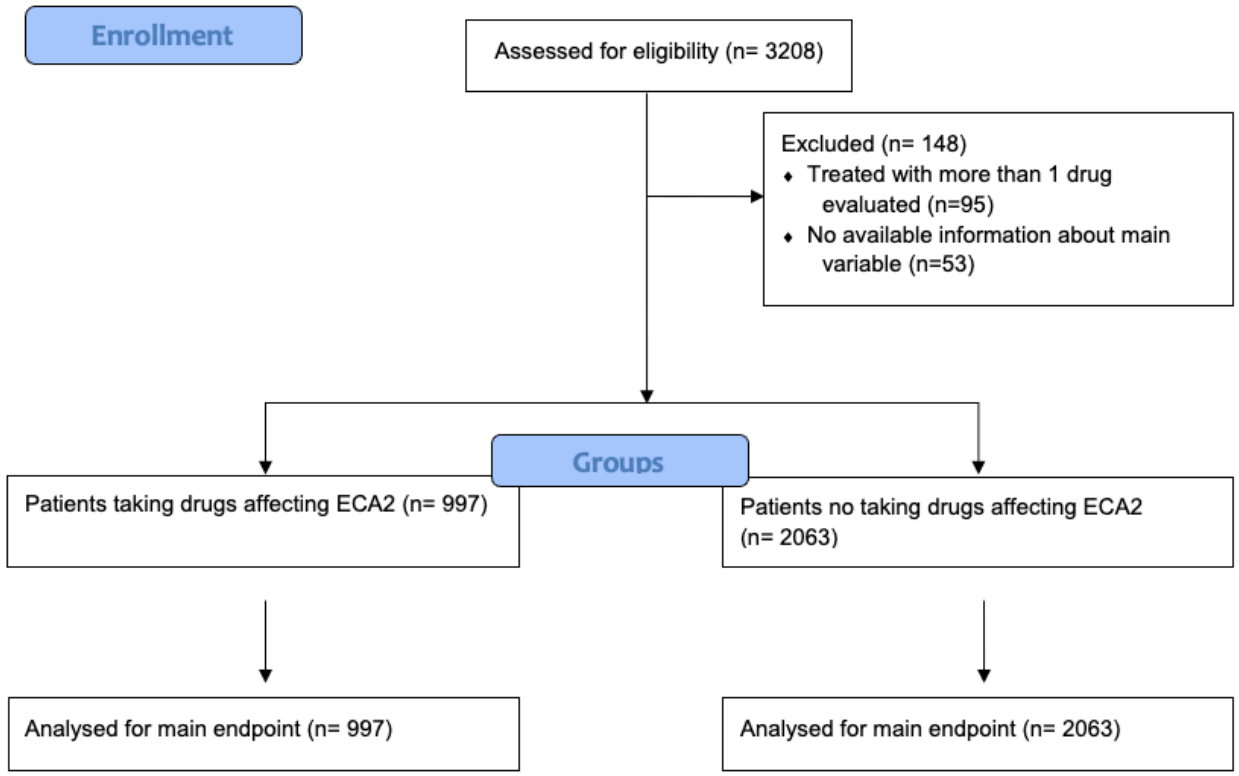
In this regard, it was initially reported that patients with severe progression of COVID-19, with a history of hypertension (HT), type 2 Diabetes Mellitus (T2DM), cardiovascular disease (CVD) and chronic kidney disease (CKD), presented a high crude mortality rate[13]. Some of these patients also had in common that they used ACE inhibitors for the treatment of these comorbidities, which alerted to their potential risk in coexistence with COVID-19. Later publications report a possible protective effect due to the down-regulation of ACE2 receptors, indicating that there is no basis for discontinuing treatment with this type of drug[13,14]. Undoubtedly, there is a link between the renin-angiotensin system (RAS) and COVID-19. Given the above, our research team conducted a retrospective study to assess the impact of treatment with ACE-interacting drugs (ACE inhibitors, ARBs and/or NSAIDs) on the evolution of COVID-19.

METHODS

A retrospective cohort study was carried out including all adult patients with a diagnosis of SARS-CoV-2 infection confirmed by PCR (Polymerase Chain Reaction), between February and May of 2020, in the region of Alava (Spain). The study was approved by the Basque Research Ethics Committee on the 21 of April 2020.

Data source was the electronic medical record, and they were provided by the Health information Subdirectorate of Health Public Service of Basque Country (Osakidetza).

Figure 1. Flow Diagram



The main outcome variable evaluated was the poor disease progression pneumonia, acute pulmonary edema, stroke, acute respiratory syndrome and hypoxemia, cardiac involvement or thrombo-embolic phenomena). The study factor was the treatment with ACE2-overexpressing drugs (yes/no): *non-steroidal anti-inflammatory drugs (NSAIDs), angiotensin II receptor blockers (ARBs) and angiotensin-converting enzyme inhibitors (ACEi) (yes/no)*. Other variables as need for hospital admission, presence of comorbidities (hypertension, diabetes, chronic bronchitis), sex and age were also collected.

Statistical analysis

Clinical and demographic characteristics of treated and untreated patients are presented in terms of frequency and percentage as being qualitative variables. The differences between groups has been analyzed through the Chi-square test.

To answer the primary endpoint (poor evolution of COVID-19 infection), crude and adjusted predictive models were built. Confounding variables as age (cut-off 65 years), gender and comorbidities (HTN, chronic bronchitis and diabetes) were included in adjusted model due to the higher risk of poor evolution in patients older than 65 years, men and patients with the comorbidities described above.

The logistic regression model only included as valid cases those patients with data on all variables included in the study. The confidence level was set at 95%. Statistical analysis was carried out with the statistical software IBM SPSS version 23.0 and R version 3.5.0.

Table 1. Clinical and demographic characteristics of exposure and no exposure patients (N= 3060)

		Treated with ACE2-overexpressing drugs		
		No (N=2063)	Yes (N=997)	p-value
Age (n;%)	≥65 years old	702 (34.0%)	534 (53.6%)	<0.001
Genre (n; %)	Men	860 (41.7%)	423 (42.4%)	0.726
Diabetes (n;%)	Yes	197 (9.55%)	212 (21.3%)	<0.001
Hypertension (n; %)	Yes	298 (14.4%)	507 (50.9%)	<0.001
Chronic bronchitis (n; %)	Yes	33 (1.60%)	38 (3.81%)	<0.001

Table 2. Clinical and demographic characteristics by subgroup (N= 3060)

Variables		IECAs (N=309)	ARBs (N=279)	NSAIDs (N=409)	No Users (N=2063)
Age (n;%)	≥65 years old	228 (73.8%)	211 (75.6%)	95 (23.2%)	702 (34.0%)
Genre (n;%)	Men	168 (54.4%)	146 (52.3%)	109 (26.7%)	860 (41.7%)
Diabetes (n;%)	Yes	92 (29.8%)	98 (35.1%)	22 (5.4%)	197 (9.55%)
Hypertension (n;%)	Yes	235 (76.1%)	225 (80.6%)	47 (11.5%)	298 (14.4%)
Chronic Bronchitis (n;%)	Yes	16 (5.2%)	13 (4.7%)	9 (2.2%)	33 (1.60%)

Table 3. Frecuency of poor evolution by therapeutic subgroup

	Poor evolution	Good evolution
IECAi	117 (17.9%)	192 (11.2%)
ABRs	104 (16.3%)	175 (10.3%)
NSAIDs	121 (18.4%)	288 (15.9%)

RESULTS

A total of 3208 adult patients with a PCR-confirmed diagnosis of SARS-CoV-2 infection were identified, but only 3060 were analyzed for the main outcome variable because there was no information available for 53 patients (figure 1).

On the other hand, 95 patients were excluded from the analysis because they were treated concomitantly with several of the drugs evaluated in order to avoid synergistic effects between drugs, and to be able to assess the effect of each drug independently. Of the 3060 patients, 10.10% (n=309) were treated with ACEi, 9.12% (n=279) with ARBs and 13.37% (n=409) with NSAIDs. The mean age was 60.07 years (standard deviation 20.53), and 58.1% (95% CI: 56.4%-59.8%) were female. Statistically significant differences were observed in the variables age and presence of comorbidities between patients treated and no treated with ACE2-overexpressing drugs (see table 1). Clinical and demographic characteristics of patients, broken down by drug subgroup are shown in table 2.

Bad disease progression

A total of 3060 adult patients with a PCR-confirmed diagnosis of SARS-CoV-2 infection were included. The main outcome variable assessed was poor disease progression. The frequency of bad evolution is shown in the table 3, broken down by type of drug. The potentially confounding variables listed in table 2 were included in the adjusted model.

Angiotensin-converting enzyme inhibitors (ACEi)

The 10.10% (n=309) of the total number of patients included in the study were on treatment with these drugs.
In the crude model, a significant higher risk of poor COVID-19 progression was observed in patients treated with ACEi (OR; 1.740; 95% CI 1.355-2.235; p≤0.001). However, when repeating the model including confounding variables (sex, age above 65 years and presence of comorbidities (diabetes mellitus, arterial hypertension and chronic bronchitis), no significant increased risk was observed in patients treated with ACEi (OR 1.245 95% CI 0.927-1.673; p= 0.146) (table 4).

Angiotensin II receptor blockers (ARBs)

The 9.12 % (n=279) of all patients included in the study were on treatment with ARBs. In the crude model, a significant higher risk of poor COVID-19 progression was observed in patients treated with ARBs (OR 1.697; 95% CI 1.306-2.205; p≤0.001). However, when repeating the model including confounding variables (sex, age above 65 years and presence of comorbidities (diabetes mellitus, arterial hypertension and chronic bronchitis), no significant increased risk was observed in patients treated with ARBs

(OR 1.345 95% CI 0.980-1.847; p= 0.067) (table 4).

Nonsteroidal Anti-inflammatory Drugs (NSAIDs)

The 13.37% (n=409) of patients included in the study were on treatment with NSAIDs. In the crude model, no increased risk of poor COVID-19 progression was observed in patients treated with NSAIDs (OR 1.200; 95% CI 0.950-1.516; p=0.127). However, when repeating the model including confounding variables (sex, age above 65 years and presence of comorbidities (diabetes mellitus, arterial hypertension and chronic bronchitis), a higher risk of poor progression was observed (OR 1.466; 95% CI 1.149 -1.870; p= 0.002) (table 4).

DISCUSSION

Concerns about whether ARBs, ACEi and NSAIDs may have a deleterious effect on morbidity and mortality in patients with COVID-19 is based on the hypothesis that these drugs might up-regulate ACE2 in target cells, facilitating COVID-19 infection[11]. To help clarify this question, a retrospective study was conducted in 3060 patients with a confirmed diagnosis of COVID-19.

Based on the results of our analysis, NSAIDs seem to have a negative influence on the course of COVID-19 (including progression to pneumonia, acute pulmonary edema, stroke, acute respiratory syndrome and hypoxaemia, cardiac involvement and thromboembolic phenomena). No significant increased risk of poor disease progression were observed for ACEi and ARBs.

The results obtained in relation to ACEi and ARBs are in line with those reported by other research groups. In a meta-analysis of studies assessing the impact of ACEi/ARBs in the incidence of adverse severe events (ASE) defined as intensive care unit admission or the need for assisted ventilation in COVID-19 positive patients, the use of these drugs was associated with a reduction of AES risk (OR 0.68; 95% CI, 0.53-0.88; p<0.001)[15]. In a prospective randomized open label trial, which compared continuation versus discontinuation of renin-angiotensin system inhibitors in patients admitted to hospital with COVID-19, authors concluded that renin-angiotensin system inhibitors can be safely continued in patients admitted to hospital with COVID-19[16]. Lopes et al, obtained similar results[17].

As discussed above, initial concerns about the use of ACE inhibitors and ARBs were based on the fact that entry of SARS-CoV-2 into cells is mediated by the ACE2 receptor[10]. Since Renin–Angiotensin–Aldosterone System (RAAS) inhibitors can increase ACE2 expression, it was hypothesized that SARS-CoV-2-infected patients were at increased risk of severe disease, as the high number of ACE2 receptors would allow the virus to enter host cells through many entry points, leading to extensive viral replication, cell death and thus triggering severe organ damage[9,10,18]. However, it has been proven that ACE2 counteracts the AT1R-mediated pro-inflammatory actions of Angiotensin II[19], and ACE inhibitors reduce thrombosis promoted by angiotensin system imbalance[19]. By breaking down angiotensin II, ACE2 has an important protective role in cardiovascular homeostasis[19] Despite initial theoretical concerns that RAAS inhibitors may be associated with serious infections and adverse events, data from large studies and meta-analyses have shown that their use is safe in patients with COVID-19 infection[19].

In relation to NSAIDs, we have found a significant increase of risk of poor progression, which don't agree with results published by other research group. In a prospective cohort study carried out by Drake et al.[20] after adjusting for explanatory variables, NSAIDs use was not associated with worse in-hospital mortality (matched OR 0·95, 95% CI 0·84–1·07; p=0·35), critical care admission (1·01, 0·87–1·17; p=0·89), requirement for invasive ventilation (0·96, 0·80–1·17; p=0·69), requirement for non-invasive ventilation (1·12, 0·96–1·32; p=0·14), requirement for oxygen (1·00, 0·89–1·12; p=0·97), or occurrence of acute kidney injury (1·08, 0·92–1·26; p=0·33)[20]. In another study comparing different clinical outcomes in COVID-19 patients treated and no treated with ibuprofen, ibuprofen use was not associated with worse clinical outcomes (received supplemental oxygen p= 0.53; mechanically ventilated p>0.95; admitted to the intensive care unit p=0.72; administration of respiratory support p>0.95)[21]. In our study, main variable is a combined variable that includes pneumonia, acute pulmonary oedema, stroke, acute respiratory syndrome and hypoxaemia, cardiac involvement or thromboembolic phenomena. That may be the reason for the non-agreement.

On the other hand, it seems clear that age > 65 years and male sex are independently associated with an increased poor disease outcome[3-5] comorbidity (diabetes, hypertension, bronchitis, and hypertension) also increases this risk. These results are in line with the evidence published to date[3-5].

The present study has, however, a number of limitations. The first limitation is the study design. Indeed, we carried out a retrospective study, which has an inherent difficulty for controlling certain biases, as well as other issues such as, under-reporting. This is a common problem in retrospective studies, but it should be borne in mind that the pandemic situation during the months of March to June 2020 made this problem even more acute. Another important limitation is that COVID-19 complications were often recorded as diagnostic impressions without an ICD-10 code, mainly due to the lack of time of the clinical staff to carry out an adequate coding before patient discharge. The incomplete coding could be relevant and might affect the results of the present study. Nonetheless, it is important to note that it would affect both groups equally (i.e., patients treated with and without drugs affecting ECA2). Due to the chaotic situation experienced during the first months of the pandemic in Alava, one of the most affected regions in Spain during the first wave of the pandemic, health records were updated at a very slow

Table 4. Poor evolution in patients treated with ACE2-overexpressing drugs**

		IECAi		ABRs		NSAIDs	
		OR	**OR***	**OR**	**OR***	**OR**	**OR***
Poor evolution	1.740 (1.355-2.235); p≤0.001	1.245 (0.927-1.673); p=0.146	1.697 (1.306-2.205); p≤0.001	*1.345 (0.980-1.847); p=0.067*	*1.200 (0.950-1.516); p=0.127*	*1.466 (1.149-1.870); p=0.002*	<0.001

pace. Additionally, there might have been an under-reporting of COVID-19-related complications, especially in non-hospitalized patients. Furthermore, other specific medication that patients may have been taking for the treatment of SARS-CoV-2 infection was not collected, which could also affect the outcome.

Another limitation that not only affects our study, but is common to all other groups, is the definition of the poor evolution of the disease, as the results may vary between the different groups depending on the definition of the variable "poor evolution".

Finally, another limitation is the impossibility of reliably knowing about treatment dropouts, which often occur without prior communication to the healthcare professional responsible for follow-up, as well as low adherence to drug treatment, especially in chronic diseases, which can be as high as 50%[22,23]. The implications of low adherence translate into negative outcomes, such as increased morbidity and mortality and additional costs for healthcare systems[24]. This low adherence may be due to different factors, including the presence of unmanageable symptoms without treatment (30%) or lack of efficacy (36%)[24]. In the case of hypertension (HTN), low levels of adherence are associated with poorer disease control and adverse outcomes such as stroke, myocardial infarction, heart failure and death[25].

Despite the above-mentioned limitations, the present study has important strengths, including: a large sample size, which covered the entire population of Alava diagnosed with COVID-19 during March to May of 2020 and the fact that we carried out subgroup analyses, i.e. a separate analysis was conducted for the ACEi and the ARBs subgroups, while most of the reviewed studies performed a joint analysis for the both.

CONCLUSIONS

It does not seem reasonable to discontinue treatment with any of the drugs evaluated in patients with SARS-CoV-2 infection, a recommendation that coincides with the findings and conclusions of other groups of researchers such as Macedo et al,[26] who suggest that the increase in ACE2 receptor expression induced by these drugs could prevent its complete neutralization by the virus, preventing a hyperinflammatory pulmonary state, so that discontinuation of treatment could potentially lead to a worsening of disease severity. In relation to NSAIDs, there is also no evidence to stop treatment, although in view of the results obtained, a close follow-up by general practitioners (GP) of patient with COVID-19 infection in treatment with NSAIDs it is recommended.

Since pain accounts for at least 22% of all primary care consultations[27] and people with chronic pain visit their GPs up to five times more often than those without[28], a community-based approach may be the most appropriate focosed on achieving a rational employment of NSAIDs.

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