Subclinical association of AST/ALT and BNP as risk factors for mortality in patients with COVID-19

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Abstract: Background. In the general population, Ast/Alt is related to BNP as a predictor of mortality and in coronavirus disease 2019 (Covid-19); Elevations in alanine aminotransferase (Alt) or aspartate aminotransferase (Ast) are associated with liver damage, Ast/Alt with mortality, and brain natriuretic peptide (BNP) identifies subclinical myocardial dysfunction. Objective. To evaluate the Ast/Alt association with subclinical BNP, and its use as prognostic risk factors, at hospital admission of patients with Covid-19.

Methods. A cohort study (240 patients with Covid-19) was carried out until the clinical outcome (improvement, death). Performing descriptive statistics, curves: operating characteristic of the receiver and Kaplan Meier, Pearson's correlation, Wilcoxon test, Cox analysis and multivariate logistic.

Results. BNP (AUC 0.77; p<0.0001) and Ast/Alt (AUC 0.67; p<0.0001) predicted mortality, in patients with Covid-19, a BNP level> 67 (HR 2.99) and Ast/Alt> 1.09 (HR 1.74) explained lower survival (p<0.0001). Ast/Alt predicted BNP> 67 (AUC 0.63, p=0.0004), with significant association (r=0.219; p<0.001); both associated with: creatinine, Ast, creatine kinase, fibrinogen, D-dimer, lactate, glomerular filtration rate. Risk factors for mortality were: BNP OR 2.203, hypertension OR 45.452, Ast/Alt OR 1.004, ferritin OR 1.002, and lymphopenia OR 0.998. There was a significant decrease in BNP (p<0.05), Ast (p<0.001) and Ast/Alt (p<0.0001) in non-survivors.

Conclusions. Ast/Alt (> 1.09) and subclinical BNP (> 67) were evaluated as risk factors for mortality at hospital admission, given their association, Ast/Alt could be a risk factor for subclinical cardiovascular disease.

Key words: Ast/Alt, BNP, Covid-19, mortality, subclinical myocardial dysfunction.

Key words: COVID-19, BNP, ALT, AST.

Introduction

Since December 2019, the coronavirus disease 2019 (Covid-19) has affected more than 200 countries [1], including ours, causing a public health problem with increasing incidence, high mortality, and greater pressure on the health care system. Acute Covid-19 infection ranges from constitutional symptoms and mild pneumonia to multiple organ failure [2]. At the cardiac level, the severe inflammatory response induced by cytokines generates subclinical diastolic dysfunction of the left ventricle, new and reversible, or triggered by exacerbation of pre-existing cardiovascular disease (CVD). This process has been documented in viral diseases such as influenza, precipitating heart damage and poor prognosis [3]. On the other hand, natriuretic peptides identify hemodynamic stress such as systolic and/or diastolic dysfunction [2], and a level of brain natriuretic peptide (BNP) <100pg/ml has been associated with severity of hypoxia [4]. Furthermore, BNP cut-off points for heart failure (HF)> 100 pg/ml (sensitivity 90%, specificity 76%), in patients who are not critically ill with pneumonia, maintain a high positive predictive value as an early indicator of dysfunction ventricular systolic [5]. Likewise, the American College of Cardiology did not mention any recommendations regarding the prognosis offered by cardiac biomarkers in patients with Covid-19 [6], so at the beginning of the pandemic its use was suggested to detect and quantify injury and/or cardiac stress, however, it was not specified when and if they should be measured as prognostic markers [5].

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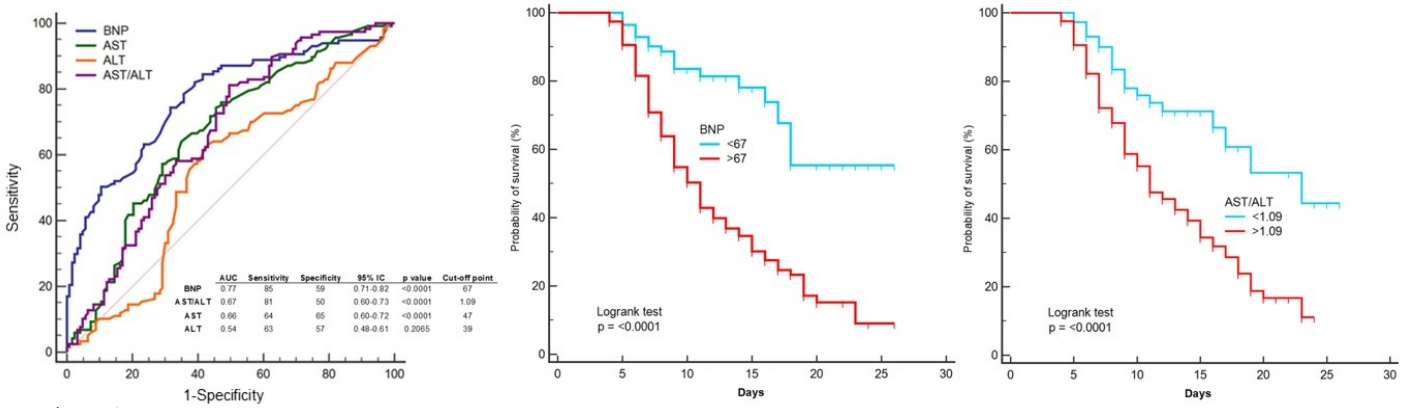

Figure 1. BNP and AST/ALT distinguish Covid-19 patients with different prognosis. (A) The ROC curve of BNP, AST/ALT, AST and ALT was used to compare the ability to predict mortality in hospitalized patients with Covid 19. Cut-off values were determined using the Youden index. The p value was obtained from the chi-square test. (B) Kaplan-Meier survival analysis in Covid-19 patients with BNP <67 and> 67, and (C) with AST/ALT <1.09 and> 1.09. *AUC:* area under the curve, *CI:* confidence interval, *BNP:* brain natriuretic peptide. *AST:* aspartate aminotransferase, *ALT:* alanine aminotransferase, *AST/ALT:* AST/ALT ratio. The p value was obtained from the Logrank test. It was considered statistically significant if p = ≤0.05.

In turn, the liver plays an important role in the course of Covid-19; since liver function tests (Lft) have modest elevations, being higher during hospitalization than at admission [7], and both elevations of alanine aminotransferase (Alt) or aspartate aminotransferase (Ast) are associated with liver damage, and in as a whole, the Ast/Alt ratio is associated with a poor prognosis including higher mortality [1]. In the general population, patients with heart disease have a greater increase in Ast, since it is located at the hepatic and myocardial level [8], and the Ast/Alt ratio (> 1.5 is associated with worsening of hepatitis acute viral and alcoholic, and increased fibrosis in liver disease) [9], is related to BNP as a predictor of all-cause mortality [8]. Given the allocation of limited resources and as the Covid-19 pandemic continues in our country, it contributes to an increase in hospitalizations and deaths, as well as the early elevation of BNP identifies greater comorbidity (such as new-onset subclinical myocardial dysfunction asso ciated with a poor prognosis) and likewise, the exploration of the Ast/Alt ratio as an indicator of poor prognosis; both parameters are useful individually to differentiate high risk of mortality, modifying crucial decision-making to improve prognosis. However, there are no studies of its possible relationship and prognosis in patients with Covid-19, so in the absence of studies and given their easy availability as low-cost and economic tools in situations of limited resources such as the one that we are living. The objective of this study was to evaluate the association of Ast/Alt with subclinical BNP and its use as prognostic risk factors, at hospital admission of patients with Covid-19.

Methods

We conducted a cohort study: prospective longitudinal observational analytical, in the Covid-19 area of Internal Medicine, in hospitals of the Secretary of Health of Mexico City (SEDESA), from May to December 2020, included data from 240 patients with infection by Covid-19 confirmed by polymerase chain reaction with reverse transcriptase by nasopharyngeal swab; approved by the research and ethics committee of SEDESA; representing minimal risk, with prior signed informed consent by the patient or responsible family member, and respecting the recommendations of the Helsinki Declaration of the World Medical Association.

At hospital admission (<24 hours), the following were recorded: epidemiological, demographic, clinical and laboratory data (Table A.1). The Ast/Alt ratio: Ast/Alt and other biochemical indicators were calculated (Eq. A.1). The patients were followed until discharge (survivor) or death (non-survivor) and recorded: last determination of BNP, Ast and Alt, days of hospital stay and need for invasive mechanical ventilation (IMV). Not included: mild cases of Covid-19, ambulatory or from intensive care units, who requested voluntary discharge, with incomplete clinical records, pregnant women, in lactation stage and/or puerperium, and since they can affect BNP levels, Ast and/or Alt, patients with: history or recent consumption of alcohol, history or diagnosis of heart disease, liver disease, chronic kidney disease, thyroid disease, chronic glucocorticoid treatment> 3 months, and in-hospital treatment with: macrolides, lopinavir/ritonavir, hydroxychloroquine, and/or tocilizumab.

Statistical analysis. To determine the data distribution, the Kolmogorov Smirnov test was performed. Quantitative variables were represented with median and interquartile range ([IQR]: 25th–75th percentile), and qualitative variables with frequency and percentage. Qualitative variables were compared with the chi-square test and quantitative variables with the non-parametric Mann-Whitney U test. The cut-off points to predict mortality and the ability of Ast/Alt to predict BNP were calculated according to the area under the curve (AUC) of the receptor operating characteristic (ROC), considering the optimal cut-off

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Biochemical parameter		Not adjusted			Adjusted		
		HR	95% CI	p value	HR	95% CI	p value
BNP	<67	0.31	0.18 – 0.52	<0.0001	0.33*	0.20 – 0.55	<0.0001
	>67	3.17	1.91 – 5.28		2.99*	1.80 – 4.99	
AST/ALT	>1.09	0.51	0.32 – 0.82	0.0059	0.57*	0.35 – 0.92	0.0214
	>1.09	1.93	1.20 – 3.09		1.74*	1.08 – 2.79	

Table 1. Prediction of mortality in patients with Covid-19. *CI:* confidence interval, *HR:* Hazard ratio, *BNP*: brain natriuretic peptide. *AST:* aspartate aminotransferase, *ALT:* alanine aminotransferase, *AST/ALT:* AST/ALT ratio. * Adjusted for age and gender. The p value was obtained from the Cox proportional hazards regression analysis. It was considered statistically significant if p = ≤0.05

point using the index of Youden. Patients were compared according to these cut-off points. Survival was analyzed using Kaplan Meier curves (KM), and groups were compared with log-rank tests. To predict mortality, the Hazard Ratio (HR) was estimated, with 95% confidence intervals (95% CI), using multivariate Cox proportional hazards regression analysis, adjusted for age and gender. Pearson's correlation was performed between BNP, Ast/Alt, and biochemical variables. To predict independent risk factors for BNP, Ast/Alt, mortality and IMV, the Odds Ratio (OR) was estimated, with 95% CI, using univariate logistic regression analysis and the variables with statistical significance were introduced to an analysis of multivariate logistic regression, and using Cox proportional hazards regression analysis. Finally, BNP, Ast/Alt, Alt and Ast were compared at hospital admission and prior to clinical outcome with the Wilcoxon paired test. Statistical analyzes of quantitative and qualitative variables were performed with SPSS software, version 25; and graphs (correlation analysis, ROC curves, survival) with R version 4.0.3. Statistically significant differences were considered when the "p" value was <0.05.

Results

Comparison of demographic and biochemical characteristics. 146 men (60.8%) and 94 women (39.2%) were included. Compared with survivors, 118 patients were non-survivors (49.2%); were older (median 63, IQR 55-69; vs 50 years), 41.5% had systemic arterial hypertension [SAH] (vs 22.1%), and 74.6% with IMV (vs 13.9%), fewer days hospitalized (median of 8, IQR 6-11; vs 9 days), at hospital admission they had higher: Ast/Alt (median of 1.36, IQR 1.11-1.62; vs 1.10), Ast (median of 56.0, IQR 39.3-80.0; vs 38.2), and BNP (median 113, IQR 80-183; vs 55); and at discharge, higher BNP (median 79, IQR 49-180; vs. 43.5), with statistically significant differences, significant differences were also observed with respect to other laboratory tests (Table A.2 and A.3).

Mortality was predicted according to the analysis of the ROC curves by calculating the AUC (Fig. 1A). BNP, Ast/Alt and Ast predicted mortality in hospitalized patients with Covid-19, compared to Ast (AUC 0.66; sensitivity 64%; specificity 65%; 95% CI 0.60-0.72; p<0.0001) or Alt, BNP (AUC 0.77; sensitivity 85 %; specificity 59%; 95% CI 0.71-0.82; p<0.0001) and Ast/Alt (AUC 0.67; sensitivity 81%; specificity 50%; 95% CI 0.60-0.73; p<0.0001), had higher AUC and sensitivity, with a statistically significant difference. By balancing sensitivity and specificity, the best cutoff value for BNP (67) and Ast/Alt (1.09) was determined. In the survival analysis (Fig. 1B, 1C), patients with Covid-19 at hospital admission, with BNP> 67 and Ast/Alt> 1.09, compared with BNP<67 and Ast/Alt<1.09, presented significantly poor survival (p=<0.0001), with a median of 14 days (95% CI 11-16), for both cut-off points. 50% of non-survivors, compared to survivors, for BNP> 67 showed a median of 11 days (95% CI 9-12; vs 20 days, 95% CI 17-22) and for Ast/Alt>1.09 it showed median of 11 days (95% CI 10-14: vs 23 days, 95% CI 17-23). Multivariate Cox proportional hazards regression analysis adjusted for age and gender was performed for BNP>67 (HR 2.99; 95% CI 1.80-4.99; p=<0.0001) and Ast/Alt>1.09 (HR 1.74; 95% CI 1.08-2.79; p=0.0214), a significantly higher risk of mortality was associated, compared to BNP<67 and Ast/Alt<1.09. Demonstrating that they were independent predictors of future mortality (Table 1).

Covid-19 patients with BNP>67, compared to BNP<67: were older (median 59, IQR 49-66, vs 53), 38.4% had diabetes mellitus 2 [DM2] (vs 25.8%), 55.3% with IMV (vs 24.4%), 33.3% survival (vs 80%), at hospital admission they had higher Ast/Alt (median of 1.32, IQR 1.07-1.62; vs 1.10), BNP (median of 112, IQR 89-164; vs 30.5), Ast (median 50, IQR 34-77; vs 40), Alt (median 41.5, IQR 23.1-56, vs 39); and at discharge, higher BNP (median 82, IQR 56.8-165; vs 29); likewise, patients with Ast/Alt>1.09, compared with Ast/Alt<1.09: were older (median 58, IQR 49.5-65; vs 51), 49.7% with IMV (vs 31.6%), 40.4 % survival (vs 72.2%), at hospital admission they had higher Ast/Alt (median 1.4, IQR 1.1-1.7; vs 0.9), BNP (median 97, IQR 53-144; vs 60), Ast (median 50.4, IQR 33.9-78; vs 43), lower Alt (median 34, IQR 19.3-53; vs 50); and at discharge, higher BNP (median 70.6, IQR 43-139; vs 40.3); for both groups with statistically significant differences, as well as between survivors and non-survivors, significant differences were also observed with respect to other laboratory

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Characteristic	Total, patients = 240	n	BNP			AST/ALT		
			<67 n = 90	>67 n = 150	p value	<1.09 n = 79	>1.09 n = 161	p value
Age (years) * [b]	55 (47 – 64)		53 (41-59)	59 (49-66)	<0.001	51 (43 – 62)	58 (49.5 – 65)	0.002
Gender (%) Male / Female [a]	146 (60.8) / 94 (39.2)		56 (62.2) / 34 (37.8)	90 (60) / 60 (40)	0.733	48 (60.8) / 31 (39.2)	98 (60.9) / 63 (39.1)	0.987
Weight (kg)* [b]	80 (70 – 90)		80 (70-89)	80 (69-92)	0.904	80 (72 – 94)	80 (69 – 90)	0.343
Size (meter)* [b]	1.65 (1.59 – 1.69)		1.65 (1.58-1.69)	1.65 (1.59-1.69)	0.491	1.65 (1.59 - 1.70)	1.65 (1.59 - 1.69)	0.490
BMI (kilogram / meter2) * [b]	30.4 (25.9 – 33.8)		30.6 (26.2-33.9)	30.4 (25.7-33.7)	0.820	30.8 (26.2 – 34)	30.1 (25.8 - 33.3)	0.299
Comorbidities (%)								
DM2 [a]	81 (33.8)		23 (25.8)	58 (38.4)	0.047	26 (32.9)	55 (34.2)	0.847
SAH [a]	76 (31.7)		26 (28.9)	50 (33.3)	0.474	25 (31.6)	51 (37.7)	0.996
DM2 + SAH [a]	27 (11.3)		9 (10)	18 (12)	0.635	11 (13.9)	16 (9.9)	0.075
Clinical outcome								
Days hospitalized * [b]	9 (7 -14)		9 (7-14)	9 (7-13)	0.336	9 (7 – 14)	9 (7 - 13)	0.370
IMV (%) [a]	105 (43.8)		22 (24.4)	83 (55.3)	<0.001	25 (31.6)	80 (49.7)	0.008
Survivors [a]	122 (50.8)		72 (80)	50 (33.3)	<0.001	57 (72.2)	65 (40.4)	<0.001
Non-survivors [a]	118 (49.2)		18 (20)	100 (66.7)		22 (27.8)	96 (59.6)	

Table 2. Demographic characteristics of patients with Covid-19 according to cut-off points for BNP and AST/ALT. *BMI*, body mass index; *DM2*, type 2 diabetes mellitus; *SAH*, systemic arterial hypertension; *IMV*, invasive mechanical ventilation. Values are expressed as numbers (percentages) unless otherwise indicated median (p25 - p75) *. The p value was obtained from: achi-square test, bMann-Whitney U test. It was considered statistically significant if p = ≤0.05.

tests (Table 2-3). The prediction of BNP was determined according to the analysis of the ROC curves calculating the AUC (Fig. 2). Ast/Alt predicted BNP>67 (AUC 0.63, 95% CI 0.56-0.69; p=0.0004); compared to Ast (AUC 0.58, 95% CI 0.52-0.65; p=0.0220) or Alt. Ast/Alt had higher AUC with statistically significant difference, suggesting better predictive capacity, than Ast or Alt alone.

There was an association (r=0.189, p=<0.05) in the correlation analysis, between BNP and Ast/Alt in non-survivors, and considering survivors, it was maintained (r=0.219; p=<0.001), with a statistically difference significant. Statistically significant associations, specifically for BNP and Ast/Alt, in common were with: creatinine (p<0.01), Ast (p<0.01), creatine kinase [Ck] (p<0.001), fibrinogen (p<0.001), D-Dimer [DD] (p<0.001), lactate (p≤0.05), and estimated glomerular filtration rate [eGFR] (p<0.01). Individually, they had other statistically significant associations, between BNP with: leukocytes (p<0.01), neutrophils (p<0.001), lactic dehydrogenase [LDH] (p<0.001), C-reactive protein [CRP] (p<0.001) and albumin (p<0.01); and between Ast/Alt with blood urea nitrogen [BUN] (p<0.001), total bilirubin [TB] (p<0.001), hemoglobin (p≤0.05), and platelets (p<0.01) (Fig. 3). A multivariate logistic regression analysis was performed, after selecting variables with statistical significance (Table A.4), the independent risk factors for BNP>67 were: SAH (OR 43.6; 95% CI 13.039-145.761; p=<0.001) and IMV (OR 4.169; 95% IC 1.350-12.872; p=0.013), in addition to Ast/Alt, age, ferritin, Ck, albumin fibrinogen ratio (Alb/Fib), and days hospitalized, with ORs close to 1; and for Ast/Alt>1.09 they were: albumin (OR 0.622; 95% IC 0.41-0.944; p=0.026) and gamma glutamyl transpeptidase (GGT, OR 0.997; 95% IC 0.994-0.999; p=0.019). Among non-survivors (Table A.5), the independent risk factors for BNP>67 were: Ast/Alt, lymphocytes (OR 0.402; 95% IC 0.908-0.972, p=<0.001) and weight; and for Ast/Alt>1.09: alkaline phosphatase (ALP, OR 7.276; 95% IC 1.498-35.343; p=0.014) and lymphocytes (OR 0.940; 95% IC 0.908-0.972, p=<0.001). The independent risk factors obtained from the Cox proportional hazards regression analysis with statistically significant association (Table 4), for mortality and IMV in common, were: BNP (OR 2.203; 95% IC 1.128-4.300; p=0.021 and OR 1.699; 95% IC 1.043-2.766; p=0.033, respectively), and SAH (OR 45.452; 95% IC 24.000-86.700; p=0.012 and OR 44.682; 95% IC 35.560-56.140; p=0.021 respectively); individually the risk factors for mortality were: IMV (OR 2.416; 95% IC 1.454-4.016; p=<0.001), in addition to Ast/Alt, Ck, ferritin, and lymphocytes, with ORs close to 1; and for IMV they were: age, LDH, DD and systemic immune inflammation index (SII), with ORs close to 1. Finally, a paired Wilcoxon test was performed, and upon

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Biochemical parameter	Total, patients n = 240	BNP			AST/ALT		
		<67 n = 90	>67 n = 150	p value	<1.09 n = 79	>1.09 n = 161	p value
Blood							
White blood cell (10^3/uL)[b]	9.83 (7.62 - 14.60)	9.21 (6.6-12)	11.3 (8.5-14.8)	**0.004**	9.5 (7 - 11.9)	11.2 (7.7 - 15.9)	**0.009**
Neutrophil (10^3/uL)[b]	8.59 (6 - 13)	7.5 (4.9-10.2)	9.55 (6.5-13.3)	**0.004**	8 (5.5 - 10.2)	9.5 (6.2 - 13.7)	**0.007**
Lymphocyte (10^3/uL)[b]	0.90 (0.60 - 1.20)	0.91 (0.6-1.3)	0.9 (0.6-1.13)	0.232	0.9 (0.6 - 1.2)	0.9 (0.6 - 1.1)	0.643
Hemoglobin (gr/dL)[b]	14.30 (12.20 - 15.90)	14.69 (13-16)	14 (11.8-15.5)	0.076	14.6 (13 - 16.2)	14 (11.8 - 15.5)	0.067
Platelet (10^3/uL)[b]	269.000 (200.910 – 335,000)	265.500 (204.000-321.000)	269.500 (196.000-346.000)	0.927	278,000 (215.000–360.000)	264.000 (184.200 – 330.000)	0.142
HbA1c (%)[b]	7.30 (6.28 - 9.30)	7.15 (5.9-9.4)	7.44 (6.5-9.3)	0.589	7.2 (5.9 - 9.4)	7.4 (6.4 - 9.3)	0.620
Kidney							
BUN (mg/dL)[b]	22.83 (14.53 - 32)	19.2 (13-30)	24.25 (15-32)	**0.041**	21 (14.4 – 30)	24 (14.5 - 34.5)	0.183
Creatinine (mg/dL)[b]	0.90 (0.71 - 1.30)	0.89 (0.73-1.14)	0.92 (0.70-1.33)	0.527	0.8 (0.7 - 1.1)	0.9 (0.7 - 1.3)	0.085
eGFR (ml/min/1.73 m^2)[b]	88 (62 - 104)	91 (66-103)	86 (55-104)	0.388	101 (66 – 108)	85 (55 – 101)	**0.004**
Liver							
Glucose (mg/dL)[b]	142.0 (111.5 - 226.5)	133.5 (102.2-226)	150.5 (115-227)	0.250	125 (104 – 192)	161 (115 - 243.5)	**0.007**
Albumin (gr/dL)[b]	2.9 (2.4 - 3.4)	3.1 (2.7-3.6)	2.8 (2.3-3.3)	**<0.001**	3.1 (2.5 - 3.7)	2.8 (2.3 - 3.3)	**0.006**
TB (mg/dL)[b]	0.61 (0.50 - 0.90)	0.6 (0.5-0.8)	0.67 (0.5-0.9)	0.439	0.7 (0.5 - 0.9)	0.6 (0.5 - 0.9)	0.292
GGT (IU/L)[b]	124.0 (62.5 - 184.5)	121 (70-185)	124 (60-180)	0.839	139 (88 – 217)	114 (50 – 174)	**0.011**
ALP (IU/L)[b]	104.99 (72.00 - 153)	99.6 (71.3-151)	109.5 (72-154)	0.758	109 (73 – 154)	100 (72 - 148.4)	0.734
AST (IU/L)[b]	47.0 (31.0 - 74.5)	40 (24 - 57)	50 (34-77)	**0.032**	43 (26 – 59)	50.4 (33.9 – 78)	**0.016**
AST (IU/L) prior to outcome[b]	39 (25 - 57)	39.5 (24 - 57)	39 (25 - 58)	0.389	36 (25 – 52)	41 (24.5 – 60)	0.225
ALT (IU/L)[b]	41.0 (24.0 - 59)	39 (24-64)	41.5 (23.1-56)	**<0.001**	50 (33 – 71)	34 (19.3 – 53)	**<0.001**
ALT (IU/L) prior to outcome[b]	36 (24 - 56)	34.7 (19.1 - 55.6)	36 (26.0 -56.0)	0.144	37 (25 – 61)	35.7 (24 - 55.5)	0.671
Heart							
CK (IU/L)[b]	127.50 (54.15 - 285.90)	68 (38.4-155.9)	196 (86-355)	**<0.001**	84.2 (54 – 195)	165 (58 – 325)	**0.007**
CK-MB (IU/L)[b]	21.65 (14.00 - 58.50)	14.9 (12-23)	30.4 (17-78)	**<0.001**	18 (13 – 35)	23 (15 – 65)	**0.013**
BNP (pg/ml)[b]	85.0 (41.0 – 123.5)	30.5 (17.5-50)	112 (89-164)	**<0.001**	60 (28 – 95)	97 (53 – 144)	**<0.001**
BNP (pg/ml) prior to outcome[b]	63.1 (35.2 - 110)	29 (15.1 - 51)	82 (56.8 - 165)	**<0.001**	40.3 (21 - 74.8)	70.6 (43 – 139)	**<0.001**
LDH (IU/L)[b]	397.50 (279.50 - 550.09)	316.58 (237-432.19)	459.6 (335-603)	**<0.001**	369.8 (245 – 483)	422 (303 - 574.5)	**0.013**
Microcirculation dysfunction							
DD (ng/ml)[b]	1144 (542 - 2800)	766 (392-1570)	1670 (755-3380)	**<0.001**	828 (383 – 1740)	1530 (612 - 2985.5)	**<0.001**
Fibrinogen (gr/L)[b]	500 (372 - 644)	444 (333-554)	542 (398-715)	**<0.001**	445 (330 – 595)	516 (401 - 683.5)	**0.015**
Lactate (mmol/L)[b]	1.5 (1.1 - 1.9)	1.3 (1-1.8)	1.6 (1.2-2)	**0.005**	1.4 (1 - 1.9)	1.5 (1.1 – 2)	0.205
Systematic inflammation							
Ferritin (µg/L)[b]	697.50 (475.83 - 820)	669.5 (400-793.24)	706 (565.27-830)	0.122	646 (420 – 772)	702 (529.5 - 838.5)	0.061
CRP (mg/dL)[b]	17.07 (8.39 - 20.85)	13.5 (5-19.4)	18.6 (12.1-24.5)	**<0.001**	16.5 (6.8 - 19.7)	18 (9.1 - 21.9)	0.140
Procalcitonin (ng/L), n (%)	198 (82.5)	81 (90)	117 (78)		68 (86.5)	130 (80.7)	
<0.5[a]	22 (9.2)	3 (3.3)	19 (12.7)	**0.033**	6 (7.6)	16 (9.9)	0.586
2[a]	20 (8.3)	6 (6.7)	14 (9.3)		5 (6.3)	15 (9.3)	
>10[a]							
Biochemical indicators							
AST/ALT[b]	1.20 (1.02 - 1.54)	1.10 (0.89-1.36)	1.32 (1.07-1.62)	**<0.001**	0.9 (0.7 - 1.0)	1.4 (1.1 - 1.7)	**<0.001**
AST/ALT prior to outcome[b]	1.08 (0.78 - 1.51)	1.11 (0.76 - 1.52)	1.08 (0.78 - 1.48)	0.660	0.9 (0.7 - 1.4)	1.1 (0.8 - 1.5)	0.062
NLR[b]	9.90 (5.85 - 17.10)	7.63 (4.57-14)	11.43 (6.75-18.5)	**0.004**	7.6 (4.5 - 15.7)	10.7 (6.5 - 17.3)	0.091
PLR[b]	295.55 (198.81 - 438.44)	285.965 (198.888-415.000)	309.723 (198.750-473.636)	0.428	288.1 (212.3 – 450)	296.6 (193.7 - 437.3)	0.479
dNLR[b]	5.80 (3.50 - 9.25)	4.84 (3.37-7.5)	6.44 (3.71-9.92)	**0.014**	4.7 (3.4 - 8.3)	6.3 (3.5 - 10.2)	**0.039**
SII[b]	2517.49 (1191.31 - 4677)	2007.89 (1054.11 – 4204.80)	3033.22 (1409.16– 4867.80)	**0.023**	2214.3 (1212.6 - 4204.8)	2688 (1180.6 – 4719)	0.484
LIN/PCR[b]	0.05 (0.03 - 0.12)	0.071 (0.044-0.248)	0.049 (0.030-0.095)	**<0.001**	0.06 (0.03 - 0.17)	0.05 (0.03 - 0.11)	0.456
PCR/ALB[b]	5.54 (2.59 - 8.46)	4.24 (1.51-6)	6.25 (3.82-9.8)	**<0.001**	4.8 (2.2 - 6.7)	5.8 (3 - 8.8)	**0.030**
ALB/FIB[b]	0.58 (0.41 - 0.82)	0.719 (0.510-0.931)	0.523 (0.352-0.716)	**<0.001**	0.7 (0.4 - 0.9)	0.5 (0.3 - 0.7)	**<0.001**
PNI[b]	34 (28.25 - 39.50)	36 (31.5-41.5)	31.5 (27-38)	**<0.001**	35.5 (30 - 41.5)	33 (27.5 - 37.6)	**0.012**
DD/BNP[b]	16.13 (7.67 - 34.59)	26.6 (11.55-60.8)	12.33 (5.58-24.3)	**<0.001**	16.5 (7.8 - 37.6)	16.1 (7.3 - 33.7)	0.775

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Table 3. Biochemical characteristics of patients with Covid-19 according to cut-off points of BNP and AST / ALT. *HbA1c*, Glycated Hemoglobin; *BUN*, blood urea nitrogen; *eGFR*, estimated glomerular filtration rate; *TB*, total bilirubin; *GGT*, gamma glutamyl transpeptidase; *ALP*, alkaline phosphatase; *AST*, aspartate aminotransferase; *ALT*, alanine aminotransferase; *AST/ALT*, AST/ALT ratio; *LDH*, lactic dehydrogenase; *CK*, creatine kinase; *CK-MB*, creatine kinase-MB; *DD*, D-Dimer; *BNP*, brain natriuretic peptide; *CRP*, C-reactive protein; *NLR*, neutrophil lymphocyte ratio; *PLR*, platelet lymphocyte ratio; *dNLR*, lymphocyte neutrophil derivative ratio; *SII*, systemic immune inflammation index; *PNI*, prognostic nutritional index; *ALB/FIB*, albumin fibrinogen ratio; *CRP/ALB*, albumin CRP ratio; *LIN/CRP* lymphocyte CRP ratio; *DD/BNP*, DD BNP ratio. Values are expressed as median (p25 - p75) unless otherwise indicated number (percentage). The p value was obtained from: achi-square test, Mann-Whitney U test. It was considered statistically significant if p = ≤ 0.05.

discharge a decrease was found among non-survivors of: BNP (p=<0.05), Ast (p=<0.001) and Ast/Alt (p=<0.0001), with statistically significant differences compared to survivors (Fig. 4).

Discussion

The main cause of death in Covid-19 is acute respiratory distress syndrome (ARDS) [2], however, the second most affected organ is the heart [3,10], causing myocardial dysfunction [1,11], and in liver acute liver injury [1]. On the other hand, Covid-19 implies significant elevation of multiple biomarkers, specifically BNP in patients who died or critically ill [12], and Ast/Alt in non-survivors [13], being BNP a marker of cardiovascular risk, and Ast of liver and myocardial involvement due to its location [8]; these findings, according to ours, would corroborate that their elevation to hospital admission in patients with Covid-19 are risk factors that indicate a poor prognosis. Notably BNP> 67 (AUC 0.77) and Ast/Alt> 1.09 (AUC 0.67) predicted mortality, with HR of 2.99 and 1.74 respectively, explaining lower survival (11 days), similar to that reported in the general population [8], and with lower cut-off points as 28-day prognosis of death [14,15], suggesting predisposition to latent CVD [8,16], therefore, its prognostic value goes in the same direction as in other studies [1,12,14,17].

On the one hand, our country has a high prevalence of chronic diseases according to the 2018 National Health and Nutrition Survey of Mexico, 36.1% of adults> 19 years old were obese, 39.1% were overweight, 13.7% with DM2 and 25% with SAH [18]; and according to 2019 reports [19] the 5 main causes of death: ischemic heart disease, DM2, chronic kidney disease, cirrhosis [7] and cerebrovascular accident; contrasting with world reports of factors associated with death from Covid-19, such as: male gender, age> 65 years, ethnicity, SAH, DM2, CVD, and respiratory disease [20], while noting factors such as limited resources and absenteeism due to infections in health workers [12,21]. Although we did not find gender differences, we were able to identify other factors previously indicated such as: older age, SAH, IMV, days hospitalized between non-survivors and survivors, similar to what was reported [1,14,15,20,22,23]. Similarly, when comparing patients with BNP> 67 and Ast/Alt> 1.09 at hospital admission, we found older age, IMV and lower survival, and curiously DM2 as comorbidity, different from what was observed [8,21,23,24] , where the prevalence of chronic diseases in our country could explain it. In addition, upon admission to hospital, we observed laboratory parameters between non-survivors and survivors with a significant difference, specifically BNP and Ast/Alt were high, coinciding with patients with BNP> 67 and Ast/Alt> 1.09, with values close to those of non-survivors, similar to other reports [1,14,15]. Over time, the significant decrease in BNP, Ast and Ast/Alt, among non-survivors, was similar to that reported in the general population [8], contrary to that reported in Covid-19 [1]. The "bystander hepatitis" described as an increase in transaminases due to immune stimulation or systemic inflammation, due to viral infections without compromising liver function [10], a similar effect on BNP could be caused by Covid-19 without compromising cardiac function, therefore further study is required.

In the cardio-hepatic interaction, Ast/Alt (AUC 0.63) predominates in the prediction of BNP> 67, with a significant correlation (r=0.219, p=<0.001), similar to that established in the general population [8]. When evaluating its association with other clinical and biochemical data independently related to higher mortality and severity [3,4,25], its association was constant with BNP and Ast/Alt in common and individually, such as: Ck, Ast, fibrinogen, creatinine, eGFR, DD, and lactate. In addition, for BNP with: CRP, leukocytes, neutrophils, and albumin [1,3,7,19,25]; similarly, Ast/Alt with: BUN, Alt, TB, hemoglobin and platelets [1,8,25], indicating a systemic inflammatory response aggravated by the storm and release of pro-inflammatory cytokines [10], higher degree of hypoxia [4], lung damage and death from ARDS [3,25], revealing the need for timely hospitalization. Interestingly, in patients with Ast/Alt> 1.09 we observed older age and lower Alt, similar to that reported in aging, frailty, and death in old age [8,25].

Complete knowledge of the factors related to BNP> 67 and Ast/Alt> 1.09 was essential since their early identification is key in patients who require timely care, such as: SAH and IMV mainly, although other risk factors with association statistically significant, they had OR close to 1, our results are

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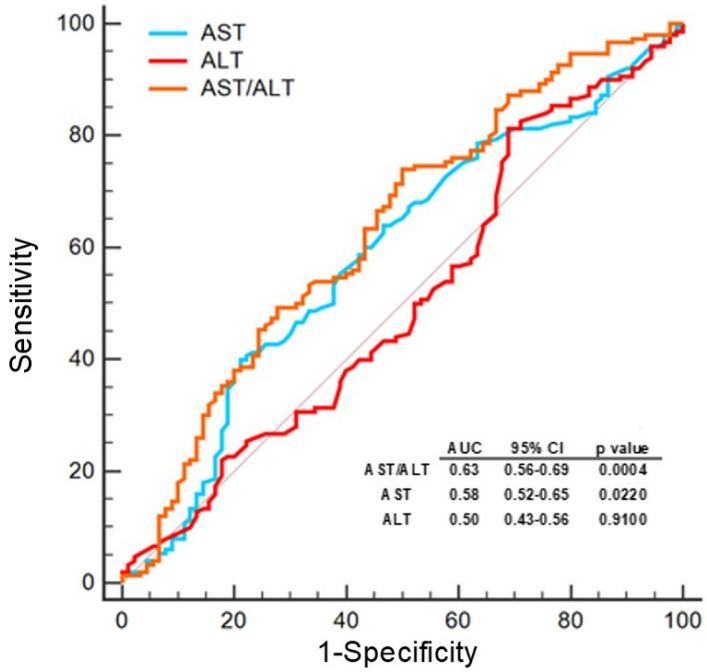

Figure. 2. AST/ALT predicts BNP level in hospitalized patients with Covid-19. The ROC curve of AST/ALT, AST and ALT was used to compare the predictive capacity of BNP> 67. *AUC*: area under the curve, *CI*: confidence interval, *AST*: aspartate aminotransferase, *ALT*: alanine aminotransferase, *AST/ALT*: AST/ALT ratio. The p value was obtained from the chi square test. It was considered statistically significant if p = ≤0.05.

similar with reports where they are associated with greater severity and mortality [3,4,25], for BNP> 67: age, Ck, ferritin, Alb/Fib, days hospitalized [3,10,22,23,25], specifically Ast/Alt is reported associated with the probability of suffering systemic injury, including the circulatory system and the heart, deteriorating myocardial function [8]; and for Ast/Alt> 1.09: GGT and albumin associated with delay in viral elimination of Covid-19, greater severity and death [25]. Among non-survivors, lymphopenia was associated as an independent risk factor, compared with critically ill patients due to Covid-19, who also present an excessive immune and inflammatory response, due to a decrease in the regulatory function of T cells [3,7,25]; and ALP indicating greater cellular damage in the bile duct [14]. Factors associated with a worse prognosis, such as uncontrolled SAH, generate more severity due to a negative impact on the immune system [21,24], and BNP associated with greater intubation with a value >100pg/ml [4,15], suggesting

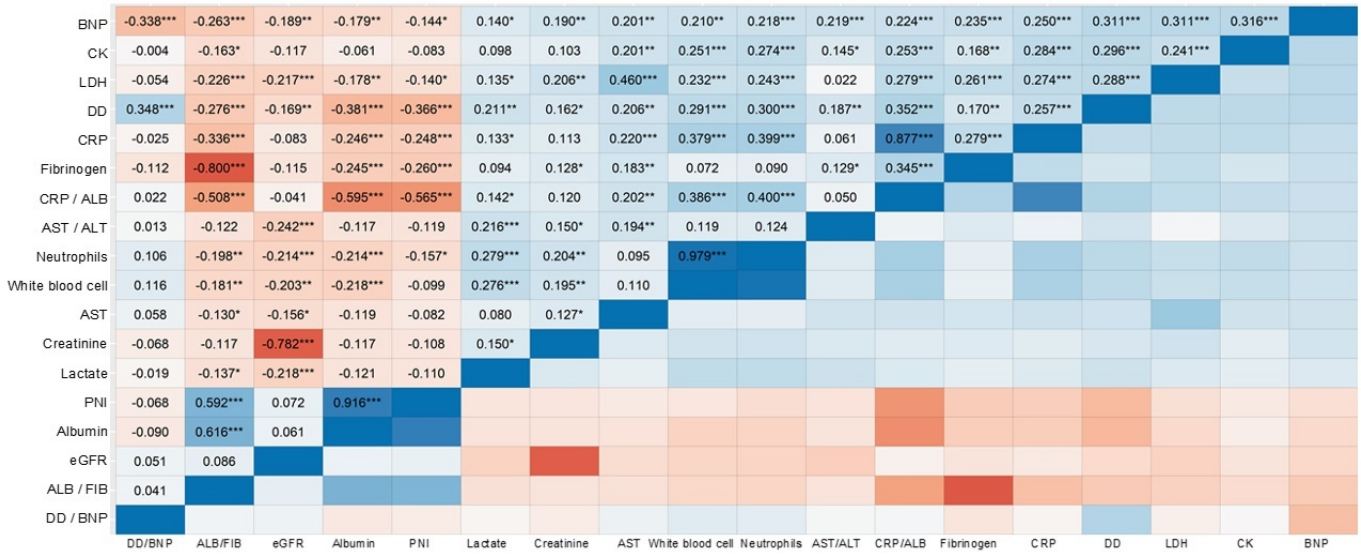

Figure 3. Association between BNP, AST/ALT and biochemical parameters in patients with Covid-19 at hospital admission. *AST*, aspartate aminotransferase; *ALT*, alanine aminotransferase; *AST/ALT*, AST/ALT ratio; *BNP*, brain natriuretic peptide; *eGFR*, estimated glomerular filtration rate; *CK*, creatine kinase; *LDH*, lactic dehydrogenase; *CRP*, C-reactive protein; *DD*, D-Dimer; *CRP/ALB*, albumin CRP ratio; *PNI*, prognostic nutritional index; *ALB/FIB*, albumin fibrinogen ratio; *DD/BNP*, DD BNP ratio. The p value was obtained from Pearson's correlation coefficient and was considered statistically significant if * p = ≤0.05; ** p = <0.01, *** p = <0.001.

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	Mortality risk				IMV risk		
	OR	95% CI	p value		OR	95% CI	p value
Age	1.022	0.998-1.047	0.067	Age	1.035	1.010-1.061	0.007
SAH	45.452	24.000-86.700	0.012	SAH	44.682	35.560-56.140	0.021
IMV	2.416	1.454-4.016	<0.001	Neutrophil	0.687	0.397-1.187	0.178
Lymphocyte	0.998	0.997-0.999	0.029	Hemoglobin	0.924	0.834-1.024	0.130
Creatinine	0.652	0.366-1.162	0.147	Glucose	1.002	0.999-1.004	0.192
CK	1.006	1.001-1.011	0.020	BNP	1.699	1.043-2.766	0.033
BNP	2.203	1.128-4.300	0.021	LDH	1.002	1.001-1.003	0.040
Fibrinogen	1.001	0.999-1.004	0.230	D-Dimer	1.003	1.002-1.004	0.039
Ferritin	1.002	1.001-1.003	0.023	CRP	1.063	0.968-1.166	0.200
CRP	1.054	0.964-1.152	0.247	AST/ALT	1.398	0.742-2.634	0.300
AST/ALT	1.004	1.001-1.006	0.042	SII	1.002	1.001 -1.003	0.020

Table 4. Cox regression analysis. Risk factors for mortality and IMV in patients with Covid-19. *OR*, odds ratio; *CI*, confidence intervals; *SAH*, systemic arterial hypertension; *IMV*, invasive mechanical ventilation; *AST*, aspartate aminotransferase; *ALT*, alanine aminotransferase; *AST/ALT*, AST/ALT ratio; *CK*, creatine kinase; *LDH*, lactic dehydrogenase; *BNP*, brain natriuretic peptide; *CRP*, C-reactive protein; *SII*, systemic immune inflammation index. It was considered statistically significant if p = ≤0.05

underlying pre-existing conditions without prior history, such as HF and atrial fibrillation [16]; other factors associated with a higher risk of death, described in several studies [1,3,14,25], were: IMV, Ast/Alt, Ck, ferritin, and lymphopenia; similarly, for IMV were: age, LDH, DD and SII [3,7,19,22,25], although BNP alone indicates hypoxic susceptibility due to endothelial dysfunction, reported in subclinical states such as prediabetes and subclinical pulmonary dysfunction [4,17], this effect is reinforced by constant associations such as Ast/Alt and other previously described indicators of severity, IMV, and death.

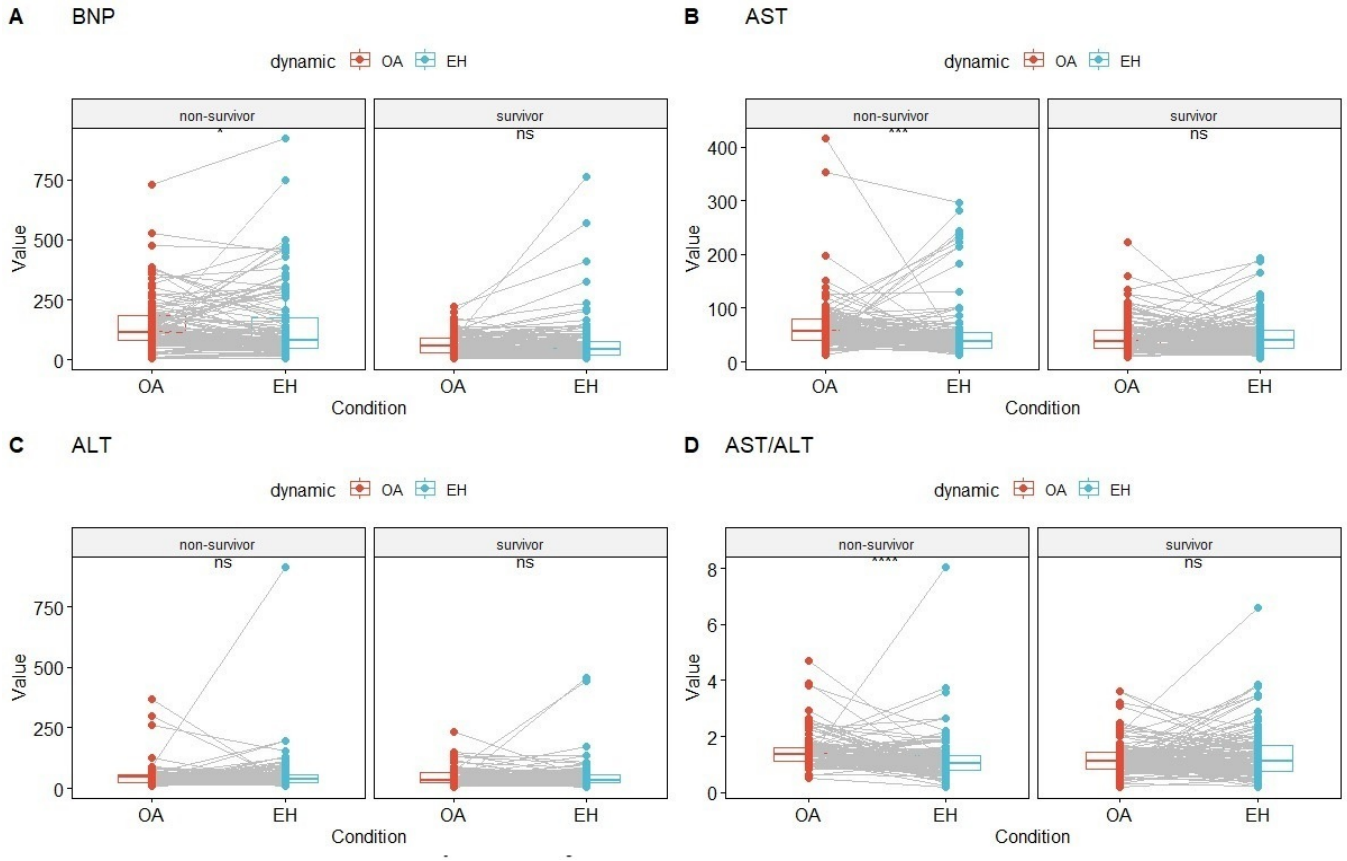

Figure 4. Comparison of dynamic changes in survivors and non-survivors, with Covid-19 during their hospitalization. (A) BNP (B) AST (C) ALT (D) and AST/ALT. *AST*, aspartate aminotransferase; *ALT*, alanine aminotransferase; *AST/ALT*, AST/ALT ratio; *BNP*, brain natriuretic peptide; *OA*, Hospital admission; *EH*, End of hospitalization; ns, not significant. Data are shown as median. The p value was obtained from Wilcoxon tests and was considered statistically significant if * p = ≤0.05; *** p = <0.001, **** p = <0.0001.

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All this information indicates that Covid-19 generates damage even with a lower level of BNP and Ast/Alt, while still causing poor prognosis, the Ast/Alt ratio, for unclear reasons, has extrahepatic implications to predict bad results as in: pancreatic cancer, cardiac injury in Kawasaki disease [1], and progression to severe pneumonia in Covid-19, the role of which remains to be further analyzed [7]; and at the cardiac level, the endothelial dysfunction and myocardial hypoxia leads to frank ventricular dysfunction and HF [10], however BNP is reported as an indicator of reversible subclinical ventricular diastolic dysfunction [3], without existing consensus on its treatment or reports of points cut-off for identification, supporting their study [7,15,18]. To our knowledge, our study is the first to address the Ast/Alt association with BNP at a subclinical level and its prognostic impact during the pandemic, so we could be underestimating latent CVD, which precedes a state of microcirculatory dysfunction [4], and in turn a subclinical state of ventricular dysfunction [3,4]; therefore, Ast/Alt could be a biomarker for its identification, in the presence of "normal" levels of BNP, leaving as a perspective, the usefulness of Ast/Alt as a risk factor for cardiovascular mortality in patients with Covid-19.

Study limitations. There is no biochemical information before the Covid-19 infection, the treatment and its relationship with the prognosis were not evaluated (it is worth mentioning that the treatments were even less standardized than now), no serum troponin levels were obtained at hospital admission (limited by scarce resources), no electrocardiogram, echocardiogram and liver ultrasound were performed, limiting the ability to identify the true burden of CVD and liver. Although these data cannot be generalized since they only represent a part of hospitalized patients at SEDESA, our results are interesting and could contribute to improving the evaluation of patients prior to hospital admission.

Conclusion

In conclusion, this study evaluated Ast/Alt (> 1.09) and subclinical BNP (> 67), as risk factors for mortality and IMV, at hospital admission of patients with Covid-19, and given their association, Ast/Alt it could be considered at hospital admission as a risk factor for subclinical CVD.

Conflicts of interests

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Appendix

Estimated glomerular filtration rate (eGFR)	Was determined using the Chronic Kidney Disease Epidemiology Collaboration formula,
NLR (neutrophil lymphocyte ratio)	Absolute neutrophils / Absolute lymphocytes
PLR (platelet lymphocyte ratio)	Absolute platelets /Absolute lymphocytes
dNLR (derived from neutrophil lymphocyte ratio)	Absolute neutrophils / (Absolute leukocytes-neutrophils)
SII (systemic immune inflammation index)	(Absolute platelets x Absolute neutrophils) / Absolute lymphocytes
LIN/CRP (lymphocyte PCR ratio)	Absolute lymphocytes / CRP
CRP/ALB (albumin CRP ratio)	CRP / Albumin
ALB/FIB (albumin fibrinogen ratio)	Albumin / Fibrinogen
PNI (prognostic nutritional index)	(Albumin x 10) + (Absolute lymphocytes x 5)
DD/BNP (DD BNP ratio)	DD / BNP

Eq. A.1 Biochemical indicators calculated. *CRP*, C-reactive protein; *BNP*, brain natriuretic peptide; *DD*, D-dimer.

Complete blood count (white blood cell, neutrophil, lymphocyte, hemoglobin, platelet)
Blood chemistry (glucose, creatinine, blood urea nitrogen)
Liver function tests (albumin, total bilirubin, gamma glutamyl transpeptidase, alkaline phosphatase, aspartate aminotransferase, alanine aminotransferase)
Cardiopulmonary profile (brain natriuretic peptide, lactic dehydrogenase, creatine kinase, creatine kinase-MB)
Microcirculation dysfunction (D-Dimer, lactate)
Inflammatory markers (ferritin, C-reactive protein, procalcitonin, fibrinogen)
Glycated hemoglobin (HbA1c)

Table A.1 Laboratory tests at hospital admission

Characteristic	Total, patients n = 240	Survivors n = 122	Non-survivors n = 118	p value
Age (years) * [b]	55 (47 – 64)	50 (40 - 55)	63 (55 - 69)	<0.001
Gender (%) Male / Female [a]	146 (60.8) / 94 (39.2)	72 (59) /50 (41)	74 (62.7) / 44 (37.3)	0.558
Weight (kg)* [b]	80 (70 – 90)	80 (68 – 90)	82 (72 - 92)	0.120
Size (meter)* [b]	1.65 (1.59 – 1.69)	1.65 (1.59 – 1.70)	1.65 (1.60 - 1.69)	0.924
BMI (kilogram / meter2) * [b]	30.4 (25.9 – 33.8)	29.8 (25.3 – 33.7)	30.8 (26.5 – 34.1)	0.180
Comorbidities (%)				
DM2 [a]	81 (33.8)	42 (34.4)	39 (33.1)	0.822
SAH [a]	76 (31.7)	27 (22.1)	49 (41.5)	<0.001
DM2 + SAH [a]	27 (11.3)	12 (9.8)	15 (12.7)	0.911
Clinical outcome				
Days hospitalized * [b]	9 (7 -14)	9 (7 – 14)	8 (6 – 11)	0.015
IMV (%) [a]	105 (43.8)	17 (13.9)	88 (74.6)	<0.001

Table A.2 Comparison of demographic characteristics of patients with Covid-19. *BMI*, body mass index; *DM2*, type 2 diabetes mellitus; *SAH*, systemic arterial hypertension; *IMV*, invasive mechanical ventilation. Values are expressed as numbers (percentages) unless

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Biochemical parameter	Total, patients n = 240	Survivors n = 122	Non-survivors n = 118	p value
Blood				
White blood cell (10^3/uL) [b]	9.83 (7.62 - 14.60)	8.45 (6.30 - 11.30)	12.95 (9.43 - 16.50)	**<0.001**
Neutrophil (10^3/uL) [b]	8.59 (6 - 13)	6.70 (4.75 - 9.77)	11.20 (8.00 - 14.80)	**<0.001**
Lymphocyte (10^3/uL) [b]	0.90 (0.60 - 1.20)	1.0 (0.70 - 1.27)	0.80 (0.60 - 1.10)	**0.035**
Hemoglobin (gr/dL) [b]	14.30 (12.20 - 15.90)	15.00 (13 - 16.30)	13.92 (11.50 - 15.00)	**<0.001**
Platelet (10^3/uL) [b]	269.000 (200.910 – 335,000)	253.500 (180,000 - 321,000)	279.500 (208,000 – 346,000)	0.131
HbA1c (%) [b]	7.30 (6.28 - 9.30)	7.08 (5.85 - 9.15)	7.49 (6.52 - 9.30)	0.276
Kidney				
BUN (mg/dL) [b]	22.83 (14.53 - 32)	17.89 (13 - 25)	28.00 (20 - 36.10)	**<0.001**
Creatinine (mg/dL) [b]	0.90 (0.71 - 1.30)	0.86 (0.70 - 1.10)	0.98 (0.78 - 1.42)	**0.007**
eGFR (ml/min/1.73 m^2) [b]	88 (62 - 104)	97 (66 - 106)	83 (49 - 100)	**0.003**
Liver				
Glucose (mg/dL) [b]	142.0 (111.5 - 226.5)	124.0 (103.0 - 185.7)	176.7 (118.0 - 250.0)	**<0.001**
Albumin (gr/dL) [b]	2.9 (2.4 - 3.4)	3.2 (2.8 - 3.7)	2.5 (2.1 - 3.19)	**<0.001**
TB (mg/dL) [b]	0.61 (0.50 - 0.90)	0.60 (0.50 - 0.80)	0.70 (0.50 - 0.90)	**0.024**
GGT (IU/L) [b]	124.0 (62.5 - 184.5)	100.5 (55.0 - 171.0)	143.0 (80.0 - 187.0)	**0.009**
ALP (IU/L) [b]	104.99 (72.00 - 153)	87.00 (69.00 - 136.00)	124.00 (82.47 - 162.00)	**0.003**
AST (IU/L) [b]	47.0 (31.0 - 74.5)	38.2 (25.0 - 59.0)	56.0 (39.3 - 80.0)	**<0.001**
AST (IU/L) prior to outcome [b]	39 (25 - 57)	39.5 (25 - 60)	38.5 (25 - 55)	0.918
ALT (IU/L) [b]	41.0 (24.0 - 59)	34.0 (23.1 - 64.0)	45.0 (26.0 - 57.0)	0.211
ALT (IU/L) prior to outcome [b]	36 (24 - 56)	34.5 (23.3 - 55)	37.9 (26 -58)	0.118
Heart				
CK (IU/L) [b]	127.50 (54.15 - 285.90)	61.60 (38.00 - 117.62)	246.50 (156.42 - 402.97)	**<0.001**
CK-MB (IU/L) [b]	21.65 (14.00 - 58.50)	15.45 (12.00 - 21.00)	51.50 (22.90 - 87.00)	**<0.001**
BNP (pg/ml) [b]	85.0 (41.0 – 123.5)	55.0 (27.0 - 90.0)	113.0 (80.0 - 183.0)	**<0.001**
BNP (pg/ml) prior to outcome [b]	63.1 (35.2 - 110)	43.5 (21 - 76.4)	79 (49 - 180)	**<0.001**
LDH (IU/L) [b]	397.50 (279.50 -550.09)	299 (234 - 414.74)	501.11 (387.00 - 626.00)	**<0.001**
Microcirculation dysfunction				
DD (ng/ml) [b]	1144 (542 - 2800)	737 (379 - 1260	1940 (1100 - 3450)	**<0.001**
Fibrinogen (gr/L) [b]	500 (372 - 644)	409 (316 - 546)	573 (469 - 758)	**<0.001**
Lactate (mmol/L) [b]	1.5 (1.1 - 1.9)	1.3 (1.0 - 1.8)	1.7 (1.3 - 2.0)	**<0.001**
Systematic inflammation				
Ferritin (µg/L) [b]	697.50 (475.83 - 820)	602.50 (370.80 - 738)	749.50 (654.00 - 863.00)	**<0.001**
CRP (mg/dL) [b]	17.07 (8.39 - 20.85)	12.75 (5.76 - 18.60)	19.37 (13.90 - 26.00)	**<0.001**
Procalcitonin (ng/L), n (%)				0.133
<0.5 [a]	198 (82.5)	106 (86.9)	92 (78)	
2 [a]	22 (9.2)	7 (5.7)	15 (12.7)	
>10 [a]	20 (8.3)	9 (7.4)	11 (9.3)	
Biochemical indicators				
AST/ALT [b]	1.20 (1.02 - 1.54)	1.10 (0.85 - 1.42)	1.36 (1.11 - 1.62)	**<0.001**
AST/ALT prior to outcome [b]	1.08 (0.78 - 1.51)	1.13 (0.76 - 1.69)	1.04 (0.80 - 1.32)	0.132
NLR [b]	9.90 (5.85 - 17.10)	7.17 (4.16 - 12.18))	13.16 (7.77 - 20.37)	**<0.001**
PLR [b]	295.55 (198.81 - 438.44)	271.59 (186.25 - 395.83)	327.50 (225.71 - 507.50)	**0.009**
dNLR [b]	5.80 (3.50 - 9.25)	4.71 (2.81 - 6.91)	7.18 (4.62 - 10.86)	**<0.001**
SII [b]	2517.49 (1191.31 - 4677)	1930.81 (997.82 - 3535.71)	3502.41 (1906.45 - 6627.27)	**<0.001**
LIN/PCR [b]	0.05 (0.03 - 0.12)	0.08 (0.04 - 0.20)	0.04 (0.02 - 0.07)	**<0.001**
PCR/ALB [b]	5.54 (2.59 - 8.46)	3.98 (1.82 - 5.67)	7.74 (4.91 - 11.66)	**<0.001**
ALB/FIB [b]	0.58 (0.41 - 0.82)	0.77 (0.58 - 1.00)	0.45 (0.29 - 0.58)	**<0.001**
PNI [b]	34 (28.25 - 39.50)	37.00 (32.50 - 42)	30.00 (25.00 - 35.50)	**<0.001**
DD/BNP [b]	16.13 (7.67 - 34.59)	14.86 (6.31 - 37.76)	16.69 (9.29 - 30.75)	0.539

Table A.3 Comparison of biochemical characteristics of patients with Covid-19. *HbA1c*, Glycated Hemoglobin; *BUN*, blood urea nitrogen; *eGFR*, estimated glomerular filtration rate; *TB*, total bilirubin; *GGT*, gamma glutamyl transpeptidase; *ALP*, alkaline phosphatase; *AST*, aspartate aminotransferase; *ALT*, alanine aminotransferase; *AST/ALT*, AST/ALT ratio; *LDH*, lactic dehydrogenase; *CK*, creatine kinase; *CK-MB*, creatine kinase-MB; DD, D-Dimer; *BNP*, brain natriuretic peptide; *CRP*, C-reactive protein; *NLR*, neutrophil lymphocyte ratio; *PLR*, platelet lymphocyte ratio; *dNLR*, lymphocyte neutrophil derivative ratio; *SII*, systemic immune inflammation index; *PNI*, prognostic nutritional index; *ALB/FIB*, albumin fibrinogen ratio; *CRP/ALB*, albumin CRP ratio; *LIN/CRP* lymphocyte CRP ratio; *DD/BNP*, DD BNP ratio. Values are expressed as median (p25 - p75) unless otherwise indicated number (percentage). The p value was obtained from: achi-square test, Mann-Whitney U test. It was considered statistically significant if p = ≤ 0.05.

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Parameter	BNP>67 (n=150)					
	Univariate Analysis			Multivariate Analysis		
	OR	95% CI	p value	OR	95% CI	p value
Age	1.085	1.057-1.114	**<0.001**	1.06	1.021-1.101	**0.002**
SAH	18.12	9.374-35.018	**<0.001**	43.6	13.039-145.761	**<0.001**
Days hospitalized	0.935	0.887-0.985	**0.011**	0.976	0.796-0.963	**0.006**
IMV	2.499	1.423-4.387	**<0.001**	4.169	1.350-12.872	**0.013**
AST	1.010	1.001-1.019	**0.028**			
ALT	0.987	0.979-0.996	**0.003**			
CK	1.048	1.033-1.064	**<0.001**	1.023	1.011-1.036	**<0.001**
Ferritin	1.001	1.000-1.002	**0.017**	1.001	1.000-1.002	**0.040**
AST/ALT	1.01	1.005-1.015	**<0.001**	1.009	1.004-1.014	**<0.001**
ALB/FIB	0.010	0.003-0.040	**<0.001**	0.003	0.001-0.031	**<0.001**
	AST/ALT>1.09 (n=161)					
	Univariate Analysis			Multivariate Analysis		
	OR	95% CI	p value	OR	95% CI	p value
Age	1.028	1.007-1.049	**0.009**			
IMV	2.133	1.211-3.757	**0.009**			
White blood cell	1.082	1.02-1.148	**0.008**			
Neutrophil	1.085	1.021-1.154	**0.009**			
Albumin	0.558	0.377-0.826	**0.004**	0.622	0.41-0.944	**0.026**
GGT	0.997	0.995-0.999	**0.014**	0.997	0.994-0.999	**0.019**
AST	1.014	1.005-1.022	**0.002**			
ALT	1.003	0.997-1.010	0.329			
CK	1.002	1.001-1.004	**0.007**			
BNP	1.003	0.999-1.006	0.109			

Table A.4 Logistic regression analysis for risk factors (BNP and AST/ALT) independent of hospital admission. *OR*, odds ratio; *CI*, confidence intervals; *SAH*, systemic arterial hypertension; *IMV*, invasive mechanical ventilation, *AST*, aspartate aminotransferase; *ALT*, alanine aminotransferase; AST/ALT, AST/ALT ratio; *GGT*, gamma glutamyl transpeptidase; *CK*, creatine kinase; *BNP*, brain natriuretic peptide; *ALB/FIB*, albumin fibrinogen ratio. It was considered statistically significant if p = <0.05.

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Parameter	BNP >67 (n=100)					
	Univariate Analysis			Multivariate Analysis		
	OR	95% CI	p value	OR	95% CI	p value
Weight	0.959	0.928-0.992	**0.015**	0.961	0.923-0.999	**0.046**
BMI	0.897	0.816-0.986	**0.024**			
White blood cell	0.911	0.819-1.014	0.087			
Lymphocyte	0.468	0.234-0.938	**0.032**	0.402	0.173-0.937	**0.035**
AST	1.006	0.992-1.02	0.384			
ALT	0.993	0.985-1.001	0.098			
DD	1.000	1.000-1.001	0.052			
AST/ALT	1.012	1.003-1.021	**0.007**	1.015	1.003-1.027	**0.011**
DD/BNP	0.992	0.985-0.999	**0.028**			
	AST/ALT>1.09 (n=96)					
	Univariate Analysis			Multivariate Analysis		
	OR	95% CI	p value	OR	95% CI	p value
Lymphocyte	0.946	0.918-0.974	**<0.001**	0.940	0.908-0.972	**<0.001**
Creatinine	1.816	0.741-4.455	0.192			
GGT	0.996	0.992-1.000	**0.043**			
ALP	4.505	1.101-18.424	**0.036**	7.276	1.498-35.343	**0.014**
AST	1.014	0.995-1.034	0.149			
ALT	1.001	0.990-1.012	0.926			
BNP	1.399	0.538-3.639	0.491			
Ferritin	1.000	0.999-1.001	0.634			

Table A.5 Logistic regression analysis for independent risk factors (BNP and AST/ALT) in non-survivors to hospital admission. *OR*, odds ratio; *CI*, confidence intervals; *BMI*, body mass index; *GGT*, gamma glutamyl transpeptidase; *ALP*, alkaline phosphatase; *AST*, aspartate aminotransferase; *ALT*, alanine aminotransferase; *AST/ALT*, AST/ALT ratio; *DD*, D-Dimer; *BNP*, brain natriuretic peptide; *DD/BNP*, DD BNP ratio. It was considered statistically significant if p = ≤0.05.

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