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MAJOR ARTICLE

Elevation of Liver Fibrosis Index FIB-4 Is Associated With Poor Clinical Outcomes in Patients With COVID-19

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Background. COVID-19 is a potentially severe disease caused by the recently described SARS-CoV-2. Whether liver fibrosis might be a relevant player in the natural history of COVID-19 is currently unknown. We aimed to evaluate the association between FIB-4 and the risk of progression to critical illness in middle-aged patients with COVID-19.

Methods. In this multicenter, retrospective study with prospective follow-up of 160 patients aged 35–65 years with COVID-19, FIB-4, clinical, and biochemical variables were collected at baseline. FIB-4 ≥2.67 defined patients with risk for advanced liver fibrosis.

Results. Risk for advanced fibrosis was estimated in 28.1% of patients. Patients with FIB-4 ≥2.67 more frequently required mechanical ventilation (37.8% vs 18.3%; $P = .009$). In multivariate analysis, FIB-4 ≥2.67 (odds ratio [OR], 3.41; 95% confidence interval [CI], 1.30–8.92), cardiovascular risk factors (OR, 5.05; 95% CI, 1.90–13.39), previous respiratory diseases (OR, 4.54; 95% CI, 1.36–15.10), and C-reactive protein (OR, 1.01; 95% CI, 1.01–1.02) increased significantly the risk of ICU admission. Bootstrap confirmed FIB-4 as an independent risk factor.

Conclusions. In middle-aged patients with COVID-19, FIB-4 may have a prognostic role. The link between liver fibrosis and the natural history of COVID-19 should be evaluated in future studies.

Keywords. liver fibrosis; critical illness; FIB-4; SARS-CoV-2; COVID-19.

Coronavirus disease 2019 (COVID-19) is a potentially severe disease, caused by the recently described SARS-CoV-2, with a broad spectrum of clinical manifestations, including acute respiratory distress syndrome (ARDS) and death. Most health systems have been overwhelmed due to the rapid spread of the virus and the high demand of patients for medical attention, especially intensive care requirements [1]. Approximately 5% of the patients with symptomatic COVID-19 will progress to critical illness, mostly due to the development of ARDS [2]. Patients at higher risk of ARDS or intensive care unit (ICU) admission are those with advanced age or comorbidities, including previous history of metabolic risk factors such as type 2 diabetes mellitus (T2DM) or hypertension. Alteration of liver biochemistry (ie, elevation of aspartate aminotransferase [AST]/alanine aminotransferase [ALT]) or liver function tests (ie, hyperbilirubinemia or hypoalbuminemia) are frequent findings in patients with COVID-19 and have been linked to major adverse clinical outcomes [2, 3].

Prevalence of liver fibrosis (≥ stage 2), mostly attributed to metabolic-associated fatty liver disease (MAFLD), is frequent in the general population (2.8%–5.6%) and increases significantly in high-risk populations (up to 18% in patients with T2DM) [4]. MAFLD is the most frequent cause of chronic liver disease in western countries and is closely related with the features of the metabolic syndrome and with numerous extrahepatic complications (ie, cardiovascular disease, neoplasms, chronic kidney disease, etc.). Advanced liver fibrosis is the main determinant of progression to cirrhosis, liver failure, and hepatocellular carcinoma [5, 6]. Furthermore, patients with advanced fibrosis (stages 3 and 4) present a higher risk of mortality compared with the reference population [7]. However, there is no information related to the prevalence and influence of liver fibrosis in COVID-19.

Noninvasive tests, based on routine biochemical and clinical parameters, are useful tools for the assessment of liver fibrosis and risk stratification. FIB-4 is a simple fibrosis score that has been validated in several etiologies of liver disease and was shown to be superior to other noninvasive markers of fibrosis [8, 9]. FIB-4 has been associated with extrahepatic clinical

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outcomes in patients with liver disease and, importantly, in various nonliver-related conditions such as intracerebral hemorrhage or atrial fibrillation [10, 11].

Finally, MAFLD has been reported in up to 38% of patients with COVID-19 and its presence has been associated with worse evolution of the respiratory disease [12]. We hypothesize that liver fibrosis might be a relevant player in the COVID-19 natural history. Therefore, we aimed to assess noninvasively the presence of advanced liver fibrosis in patients with COVID-19 and to evaluate the contribution of advanced fibrosis to clinical outcomes.

METHODS

Study Design and Participants

This observational study included patients with a confirmed SARS-CoV-2 infection at 5 tertiary-level hospitals in the region of Madrid from 26 February to 20 March 2020. Study inclusion was retrospective and follow-up was prospective until discharge from hospital, death, or end of follow-up. Diagnosis of the infection was based on RNA detection of SARS-CoV-2 in a nasopharyngeal swab sample by real time reverse-transcriptase polymerase chain reaction (rRT-PCR) followed by a second positive rRT-PCR confirmation in all patients. Patients were excluded for any of the following criteria: previous diagnosis of myopathy or platelet disorders, recipients of solid organ transplant, and patients treated with drugs known to produce myelotoxicity. Patients younger than 35 years or older than 65 years (the range of ages in which FIB-4 is less accurate) were also excluded [13]. In addition, patients with SARS-CoV-2 infection who were hospitalized for reasons other than COVID-19 were excluded. The study was performed in agreement with the Declaration of Helsinki and was approved as consent-waived by the Ethics and Clinical Research Committee of Hospital Universitario Puerta de Hierro.

Clinical Assessment

Demographic characteristics (age, sex, and race) and laboratory tests (ALT, AST, bilirubin, γ-glutamyl transpeptidase [GGT], lactate dehydrogenase [LDH], C-reactive protein (CRP), international normalized ratio [INR], platelets, hemoglobin, and white cell count) were recorded at the same time as SARS-CoV-2 detection at the emergency room. Additional information was recorded regarding concomitant medication, previous history of cardiovascular risk factors (T2DM, dyslipidemia, hypertension, smoking habit, and obesity), as well as other relevant past medical history, including chronic obstructive pulmonary disease, obstructive sleep apnea/hypopnea syndrome, ischemic or valvular heart disease, chronic kidney disease, chronic advanced liver disease, cancer (type of tumor and remission status), autoimmune disorders, and psychiatric or neurologic diseases.

Noninvasive Assessment of Liver Fibrosis

FIB-4 was calculated according to the following equation [age × AST (IU/L)]/[platelets ($\times 10^9$) × √ALT (IU/L)] from blood tests taken at the time of hospital admission, before starting any specific COVID-19 therapies. Previously published cutoffs were used to exclude and diagnose advanced fibrosis [14]. Specifically, a value of FIB-4 below 1.30 is considered as low risk for advanced fibrosis; a value of FIB-4 over 2.67 is considered as high risk for advanced fibrosis; and FIB-4 values between 1.30 and 2.67 are considered as intermediate risk of advanced fibrosis. In order to minimize overestimation of predicted advanced fibrosis, patients belonging to the intermediate FIB-4 category were considered negative for advanced fibrosis in the multivariate analysis (see Results section).

Outcomes

The primary endpoint was to evaluate noninvasively the proportion of patients with COVID-19 at risk for advanced liver fibrosis (FIB-4 ≥2.67). The secondary aims were (1) to evaluate baseline characteristics of patients according to FIB-4 categories (high risk vs low/intermediate risk for advanced fibrosis), and (2) to evaluate whether FIB-4 is associated with the need for mechanical ventilation (MV).

Statistical Analysis

Data were reported as the median and interquartile range (IQR) or mean and standard deviation (SD) for continuous variables, while frequency and percentage were used for discrete variables. Categorical variables were compared with χ^2 test and continuous variables with the Student *t* test. Nonparametric alternatives (Mann–Whitney *U* or Fisher exact test) were used for non-normal distributions. For secondary endpoint univariate and multivariate logistic regression analyses were performed. Independent variables with significance $P \leq .10$ in the univariate analysis, together with selected covariates based on their biologically plausible potential to act as confounders, were introduced in covariate-adjusted multivariate analyses (backward likelihood ratio regression analysis) to provide an optimal control for risk factors and confounders. To increase the robustness and assess the accuracy of the results provided by the logistic regression model we performed a bootstrap analysis. From the original cohort, 1000 different samples were generated by random selection and replacement using the conditional resampling method. This procedure provides a more reliable estimate of the coefficients of each covariate and therefore increases the internal validity of the results. Additionally, we specifically checked for the modifier effect of FIB-4 categories on the covariates by including interaction terms in the models. A significant interaction would indicate that the effect of covariates was different between FIB-4 categories. Odds ratios (OR) and their 95% confidence intervals (CI) were estimated. Values were considered statistically significant when $P < .05$. SPSS Statistics (version 19.0; IBM Corporation) was used in all analyses. This observational study was conducted in accordance with the STROBE (Strengthening the Reporting in Observational Studies) statement.

RESULTS

Baseline Characteristics of the Study Population

Between 26 February and 20 March 2020, 449 patients with a confirmed SARS-CoV-2 infection attended the participating centers. Patients meeting exclusion criteria (n = 236) and those in whom FIB-4 could not be calculated at baseline (n = 53) were excluded (Figure 1). The cohort considered for the analysis comprised 160 patients. The median length of follow-up was 29 days (IQR, 26–33 days) and no patient was lost during follow-up. The baseline characteristics of the population are given in Table 1. Briefly, the median age was 55 years (IQR, 48–60 years) with a lower proportion of women (41.3%). Overall, 39.4% of patients presented with at least 1 cardiovascular risk factor, the most frequent being obesity (37%), hypertension (20%), and smoking (19.3%). Patients with a previous diagnosis of respiratory or heart disease were 12.6% and 21.3% of the population, respectively. Previous history of liver disease was reported in 13 patients (8 MAFLD, 3 hepatitis C virus, 1 hepatitis B virus, and 1 alcohol-related liver disease). No active cases of malignancy were reported. Median FIB-4 was 1.87 (IQR, 1.34–2.90). A total of 23.8% (38/160) required MV with a median time of 7 days (IQR, 4–11 days) from hospitalization to ICU admission.

Baseline Features According to FIB-4 Categories

Forty-five patients (28.1%) showed a FIB-4 ≥2.67. As expected, individual components of FIB-4 were significantly higher in the FIB-4 ≥2.67 group. An almost significant higher prevalence of cardiovascular risk factors was noted in the group of patients with FIB-4 ≥2.67 (23 [51.1%] vs 40 [34.8%]; $P = .057$). Previous heart, lung, and renal diseases showed a balanced distribution between groups with FIB-4 <2.67 and FIB-4 ≥2.67 (Table 2). Levels of acute-phase response proteins, such as CPR, were higher in the group at risk for fibrosis (87 mg/L [SD 66] vs 76 mg/L [SD 100]; $P = .020$). Other features of systemic inflammatory response were also more pronounced in the group at risk for fibrosis, that is a lower lymphocyte count (0.9 10^9/L [SD 0.3] vs 1.1 10^9/L [SD 0.5]; $P = .001$) and higher levels of LDH (393 U/L [SD 189] vs 297 U/L [SD 142]; $P = .002$).

In the group of patients at risk for liver fibrosis, need for MV occurred more frequently (37.8% vs 18.3%; $P = .009$) and time from diagnosis of COVID-19 to ICU admission was shorter (5 [SD 4] days vs 10 [SD 5] days; $P = .05$).

To evaluate the influence of COVID-19 on FIB-4 values and its individual components we retrieved previous values of AST, ALT, and platelets in 24 (15%) patients of the cohort. These laboratory tests had been done within the previous 6 months before the diagnosis of COVID-19 as part of scheduled tests at primary care. Baseline characteristics between patients with or without available blood test were similar. AST and ALT increased significantly at the time of COVID-19 diagnosis, while platelet counts remained stable from previous

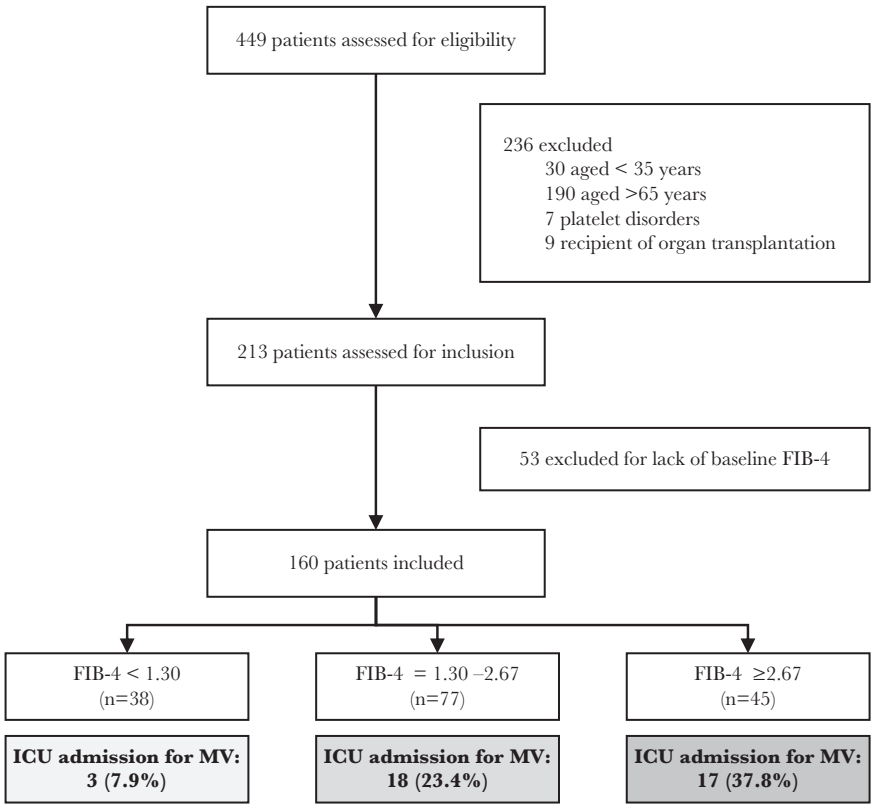

Figure 1. Flow diagram of the study. Abbreviations: FIB-4, fibrosis-4 score; ICU, intensive care unit; MV, mechanical ventilation.

Table 1. Baseline Features of the Study Population

Characteristic	Study Cohort (n = 160)
Age, y, median (IQR)	55 (48–60)
Female sex, No. (%)	66 (41.3)
Cardiovascular risk factors, any, No. (%)	63 (39.4)
T2DM, No. (%)	19 (11.9)
Hypertension, No. (%)	32 (20)
Dyslipidemia, No. (%)	26 (16.3)
Obesity, No. (%)[b]	16 (37)[b]
Current or previous smoker, No. (%)	28 (19.3)
Respiratory disease, No. (%)	20 (12.6)
Ischemic or valvular heart disease, No. (%)	34 (21.3)
Advanced chronic liver disease, No. (%)	2 (1.3)
Chronic kidney disease, No. (%)	2 (1.3)
Cancer, No. (%)[a]	9 (5.7)
Remission status, No. (%)	9 (100)
White cell count, $\times 10^9$/L, median (IQR)	5.9 (4.3–8.0)
Lymphocytes, $\times 10^9$/L, median (IQR)	1.03 (0.73–1.32)
Platelets, $\times 10^9$/L, median (IQR)	183 (154–231)
Hemoglobin, g/dL, median (IQR)	14.3 (13.3–15.3)
AST, U/L, median (IQR)	40 (26–68)
ALT, U/L, median (IQR)	36 (22–66)
GGT, U/L, median (IQR)	49 (28–104)
Total bilirubin, mg/dL, median (IQR)	0.5 (0.35–0.68)
Albumin, g/dL, median (IQR)	4.1 (3.6–4.2)
INR, median (IQR)	1.04 (0.98–1.11)
LDH, U/L, median (IQR)	284 (211–389)
C-reactive protein, mg/L, median (IQR)	47 (13–109)
FIB-4, median (IQR)	1.87 (1.34–2.90)
FIB-4 ≥ 2.67, No. (%)	45 (28.1)
ICU admission, No. (%)	38 (23.8)
Time to ICU admission, d, median (IQR)[b]	7 (4–11)
Mechanical ventilation, No. (%)	36 (22.5)
Death, No. (%)	1 (0.6)

Abbreviations: ALT, alanine aminotransferase; AST, aspartate aminotransferase; FIB-4, fibrosis index; GGT, γ-glutamyl transpeptidase; ICU, intensive care unit; INR, international normalized ratio; LDH, lactate dehydrogenase; T2DM, type 2 diabetes mellitus.

[a]Except nonmelanoma skin cancer.

[b]Data available from 1 center only.

Table 2. Baseline Features According to the FIB-4 Category

Characteristic	FIB-4 < 2.67 (n = 115)	FIB-4 ≥ 2.67 (n = 45)	*P* Value
Age, y, mean (SD)	52.4 (8.0)	56.9 (6.3)	.001
Female sex, No. (%)	50 (43.5)	16 (35.6)	.360
Cardiovascular risk factors, any, No. (%)	40 (34.8)	23 (51.1)	.057
T2DM, No. (%)	15 (13.0)	4 (8.9)	.333
Hypertension, No. (%)	19 (16.5)	13 (28.9)	.079
Dyslipidemia, No. (%)	19 (16.5)	7 (15.6)	.882
Obesity, No. (%)[b]	10 (35.7)	6 (40)	.782
Current or previous smoker, No. (%)	21 (18.3)	7 (15.5)	.800
Respiratory disease, No. (%)	13 (11.4)	7 (15.6)	.477
Ischemic or valvular heart disease, No. (%)	18 (15.8)	12 (26.7)	.114
Advanced chronic liver disease, No. (%)	0 (0)	2 (4.4)	.102
Chronic kidney disease, No. (%)	1 (0.9)	1 (2.2)	.487
Cancer, No. (%)[a]	7 (6.1)	2 (4.4)	1.000
Remission status, No. (%)	7 (6.1)	2 (4.4)	1.000
White-cell count, $\times 10^9$/L, mean (SD)	7.3 (4.3)	6.0 (3.4)	.072
Lymphocytes, $\times 10^9$/L, mean (SD)	1.1 (0.5)	0.9 (0.3)	.001
Platelets, $\times 10^9$/L, mean (SD)	220 (71)	154 (46)	.001
Hemoglobin, g/dL, mean (SD)	14.4 (1.4)	14.4 (1.5)	.973
AST, U/L, mean (SD)	43 (31)	87 (69)	.001
ALT, U/L, mean (SD)	45 (39)	57 (35)	.067
GGT, U/L, mean (SD)	91 (112)	87 (88)	.817
Total bilirubin, mg/dL, mean (SD)	0.6 (0.4)	0.6 (0.6)	.307
Albumin, g/dL, mean (SD)	4.1 (0.8)	3.8 (0.3)	.555
INR, mean (SD)	1.1 (0.1)	1.1 (0.1)	.760
LDH, U/L, mean (SD)	297 (142)	393 (189)	.002
C-reactive protein, mg/L, mean (SD)	76 (100)	87 (66)	.020
ICU admission, No. (%)	21 (18.3)	17 (37.8)	.009
Time to ICU admission, d,[b] mean (SD)	10 (5)	5 (4)	.050
Mechanical ventilation, No. (%)	19 (16.5)	17 (37.8)	.004
Exitus, No. (%)	0 (0)	1 (2.2)	.281

Abbreviations: ALT, alanine aminotransferase; AST, aspartate aminotransferase; FIB-4, fibrosis index; GGT, γ-glutamyl transpeptidase; ICU, intensive care unit; INR, international normalized ratio; LDH, lactate dehydrogenase; T2DM, type 2 diabetes mellitus.

[a]Except nonmelanoma skin cancer.

[b]Data only available from 1 center.

laboratory tests. FIB-4 categories did not change (P = .102) between the 2 time points.

FIB-4 as a Predictor of Adverse Clinical Outcomes

During hospitalization, 23.8% (38/160) of patients were transferred to ICU facilities for invasive (n = 36) or advanced noninvasive MV (n = 2). Differences in baseline characteristics between patients requiring or not MV are depicted in Supplementary Table 1. We observed that patients needing MV presented more frequently with chronic respiratory diseases (P = .018) as well as cardiovascular risk factors (hypertension (P = .041) and obesity (P = .003)). These patients also exhibited more frequently features of systemic inflammatory response, such as higher CRP (P = .001) and lower lymphocyte count (P = .003).

Univariate association between potential predictors and need for MV are shown in Table 3. Interestingly, FIB-4 categories entailed different risks of needing MV: FIB-4 <1.30 (OR, 1; reference category), FIB-4 1.30–2.67 (OR, 3.56; 95% CI, .98–12.9), and FIB-4 ≥2.67 (OR, 7.08; 95% CI, 1.88–26.6). In multivariate analysis (Table 3) the presence of at least 1 cardiovascular risk factor (OR, 5.05; 95% CI, 1.90–13.39), history of respiratory disease (OR, 4.54; 95% CI, 1.36–15.10), CRP (OR, 1.012; 95% CI, 1.006–1.017), and FIB-4 ≥2.67 (OR, 3.41; 95% CI, 1.30–8.92) were positively associated with a greater need for MV. Bootstrapping confirmed these covariates to be robust predictors of MV (bootstrap 95% CI for FIB-4, 1.20–10.79). If patients with previously known liver disease other than MAFLD (n = 5) were excluded from the analysis, FIB-4 ≥2.67 remained as a risk factor in multivariate analysis (OR, 3.25; 95% CI, 1.24–8.53).

Table 3. Variables Found as Significant Predictors for Mechanical Ventilation

	Univariate			Multivariate								
				Model 1			Model 2			Model 3		
Variable	OR	95% CI	*P* value	OR	95% CI	*P* value	OR	95% CI	*P* value	OR	95% CI	*P* value
Sex, female	0.58	.27–1.26	.168	…	…	…	…	…	…	…	…	…
Cardiovascular risk factors, yes	3.65	1.70–7.81	.001	5.05	1.90–13.39	.001	5.10	1.95–13.34	.001	5.04	1.90–13.39	.001
Hypertension, yes	2.34	1.02–5.43	.045	…	…	…	…	…	…	…	…	…
T2DM, yes	1.17	.39–3.49	.78	…	…	…	…	…	…	…	…	…
Dyslipidemia, yes	1.92	.78–4.75	.16	…	…	…	…	…	…	…	…	…
Cardiovascular disease, yes	1.8	.76–4.29	.182	…	…	…	…	…	…	…	…	…
Respiratory disease, yes	3.1	1.18–8.19	.022	4.54	1.36–15.10	.014	4.94	1.49–16.32	.009	4.54	1.36–15.1	.014
Age, y	1.03	.98–1.08	.232	…	…	…	…	…	…	…	…	…
AST, U/L	1.01	1.0–1.02	.054	…	…	…	1.006	.99–1.01	.132	1.003	.99–1.01	.549
ALT, U/L	1.01	1.0–1.02	.05	…	…	…	…	…	…	…	…	…
Platelets, $\times 10^9$/L	1.001	.99–1.01	.82	…	…	…	…	…	…	…	…	…
GGT, U/L	1.003	1–1.006	.083	…	…	…	…	…	…	…	…	…
Bilirubin, mg/dL	3.49	1.37–8.87	.009	…	…	…	…	…	…	…	…	…
LDH, U/L	1.011	1.007–1.016	.001	…	…	…	…	…	…	…	…	…
White cells, $\times 10^9$/L	1.07	.98–1.16	.131	…	…	…	…	…	…	…	…	…
Lymphocytes, $\times 10^9$/L	0.21	.07–.6	.004	…	…	…	…	…	…	…	…	…
C-reactive protein, mg/L	1.01	1.005–1.015	.001	1.012	1.006–1.017	.001	1.011	1.006–1.016	.001	1.012	1.006–1.017	.001
FIB-4 < 1.30, yes, reference	1.00	…	.011	…	…	…	…	…	…	…	…	…
FIB-4 1.30–2.67, yes	3.56	.98–12.9	.054	…	…	…	…	…	…	…	…	…
FIB-4 >2.67, yes	7.08	1.88–26.6	.001	3.41	1.30–8.92	.012	…	…	…	3.41	1.30–8.92	.012

Results are based on multivariable logistic regression. Model 2: compared with model 1, this model evaluates AST as an independent predictor for mechanical ventilation. Model 3: compared with model 1, this model evaluates AST, as well as FIB-4, as independent predictors for mechanical ventilation.

Abbreviations: ALT, alanine aminotransferase; AST, aspartate aminotransferase; CI, confidence interval; FIB-4, fibrosis index; GGT, γ-glutamyl transpeptidase; LDH, lactate dehydrogenase; OR, odds ratio; T2DM, type 2 diabetes mellitus.

We constructed 2 additional multivariate models to evaluate specifically the influence of AST on the endpoint. The first model included AST (instead of FIB-4 to avoid collinearity) with the aim of evaluating the effect of AST individually. The second model included both AST and FIB-4 to evaluate the influence of FIB-4 in the presence of AST as a covariate. In both multivariate models, AST was excluded confirming that AST was not independently associated with the endpoint. No significant interactions between the endpoint, FIB-4 categories, and other covariates were found.

DISCUSSION

In this study, we evaluated the association between FIB-4, a liver fibrosis index, and the risk of progression to critical illness in middle-aged patients with COVID-19. The first important result of our series is that the estimated risk of liver fibrosis by FIB-4 was greater than expected, reaching 28.1%. Furthermore, patients with higher FIB-4 were more likely to require MV. Finally, our results indicate that after adjusting for other well-known risk factors that negatively influence the natural history of COVID-19 (age, comorbidities, and markers of acute inflammation), FIB-4 ≥2.67 independently increases (OR, 3.41) the need for MV in middle-aged patients.

Our results are in line with those recently published where an association between FIB-4 categories and the risk of severe COVID-19 was found in patients with MAFLD [15]. Liver fibrosis is a strong predictor of all-cause mortality and increased liver-related morbidity (liver failure, portal hypertension, and hepatocellular carcinoma) in patients with chronic liver disease [6, 16]. In the last decades, a significant number of noninvasive tests have been developed to noninvasively assess liver fibrosis. We selected FIB-4 because it is a simple score composed of age and 3 readily available laboratory parameters (AST, ALT, and platelets), which can be obtained at the emergency room. In addition, FIB-4 cutoffs have been validated in different etiologies of liver disease. This is a convenient feature for our study because we could not assess properly the etiology of a potential underlying liver disease in all patients. Previous history of liver disease was reported in 13 patients; however, prevalence of MAFLD was presumably underestimated in our cohort in light of the high prevalence of metabolic and cardiovascular risk factors, and therefore MAFLD probably accounted for a greater proportion of patients, as previously reported [12]. Unfortunately, due to the circumstances of data acquisition, noninvasive diagnosis of liver steatosis could not be obtained in this cohort.

FIB-4 is also useful to identify patients with liver diseases who are likely to have a liver-related adverse clinical outcome [17, 18]. Importantly, FIB-4 has been shown to predict nonliver-related clinical outcomes like cardiovascular mortality or risk of atrial fibrillation in patients with MAFLD [10, 19]. Likewise, FIB-4 has been shown to predict mortality in the general population [20] and clinical outcomes in nonliver-related clinical settings [11]. It should be emphasized that the aim of our study was not to elaborate a prognostic model for the need for MV in COVID-19 but to point out the possible influence of underdiagnosed liver disease in the natural history of COVID-19. Transient elevations of transaminases have been reported during COVID-19 infection [2]. Therefore, evaluation of transaminases at the time of COVID-19 might not be representative of the pre-COVID-19 status and thus FIB-4 may not be an accurate estimator of liver fibrosis. To overcome this problem, we analyzed our data in different ways: (1) we retrieved available information on blood test done within 6 months before COVID-19 diagnosis in a relatively small number of patients (15% of the total series): at the time of COVID-19 diagnosis, AST and ALT increased significantly while platelets remained stable as compared with previous values; however, there were no significant changes in FIB-4 categories; (2) we evaluated specifically the prognostic value of isolated baseline AST: in contrast to previous reports, AST was not an independent predictor either at univariate level or when adjusted by other clinical and laboratory covariates; and (3) finally, we evaluated specifically the association between the elevation of AST (ie, AST above the upper limit of normality) and the need for MV, which identified that AST elevation was an independent risk factor. However, this assessment deserves a cautious interpretation as it incorporates collinearity in the model and complicates the interpretation of the results. Importantly, it should be emphasized that estimating the presence of fibrosis with an approach different from biochemical markers is very complex during COVID-19. Liver biopsy, the current gold standard for assessing liver fibrosis, is clearly unfeasible and probably unethical and elastography is difficult to perform.

The most intriguing finding of our study is the association between elevated FIB-4 and poor COVID-19 outcomes. Strikingly, patients classified as at risk for fibrosis required MV more frequently. In this context, it is possible to speculate that advanced fibrosis may enhance the risk for development of exacerbated inflammatory response, a characteristic finding of severe COVID-19. In fact, advanced liver disease is characterized by a persistent stimulation of immune cells by pathogen-associated molecular patterns (PAMPs) and damage-associated molecular patterns (DAMPs) that activate immune cells and upregulates the production of cytokines, chemokines, and growth factors, which are released to recruit and activate additional inflammatory cells, perpetuating a state of chronic low-grade systemic inflammation [21, 22]. A similar state of low-grade inflammation has been reported in patients with obesity and insulin resistance. In fact, serum levels of interleukin-6 (IL-6) have been correlated with the degree of obesity and with the risk of T2DM development [23]. In the setting of an acute infection, activated macrophages secrete IL-6, which is the major inducer for the synthesis of acute-phase response proteins in the hepatocyte (CRP, ferritin, complement, clotting factors, etc.). The acute-phase proteins produced by the hepatocyte have direct effector function on innate immunity therefore promoting pathogen clearance [24]. Unfortunately, we do not have information regarding IL-6 levels in this cohort to evaluate specifically the interaction between IL-6 and FIB-4. We have shown, however, that elevation of FIB-4 was associated with higher levels of CRP, suggesting that inflammatory response is aggravated in patients with higher fibrosis markers.

Although we report data from a large cohort of patients with COVID-19 from tertiary-level hospitals, there are several limitations that should be discussed. First, FIB-4 components are not liver specific and may be affected by disorders other than liver disease. To overcome this problem, we excluded patients previously diagnosed with myopathies and platelet disorders. Second, our study only included patients aged between 35 and 65 years. However, for patients younger than 35 years noninvasive assessment of liver fibrosis should be done with tests other than FIB-4 (ie, elastography) because FIB-4 may underdiagnose fibrosis. On the other hand, specificity of FIB-4 for advanced fibrosis in patients older than 65 years decreases significantly and may overestimate fibrosis [13].

Furthermore, the need for MV was an endpoint of the study. Management and decision making in the overwhelming and exceptional setting of COVID-19 was not homogeneous over time in patients with advanced age, a finding that has been reported previously [1]. However, the range of age selected in the study is not generally affected by this factor. Finally, a significant proportion of patients (48%) were categorized in the intermediate FIB-4 category, which is similar to that previously reported [25]. In this group of patients additional tests should be carried out to determine the risk of advanced fibrosis [24]. This pragmatic approach, however, could not be adopted in the setting of coronavirus. Although some patients in the grey zone would be positive for advanced fibrosis, we decided to categorize this subset of patients as negative for advanced fibrosis in multivariate analysis in order to minimize the risk of overestimation of fibrosis severity.

In conclusion, our results suggest that in middle-aged patients with COVID-19, FIB-4 may have a relevant prognostic role. Whether FIB-4 really accounts for liver fibrosis or it is just

a change induced by COVID-19 will need to be clarified in future studies.

Supplementary Data

Supplementary materials are available at *The Journal of Infectious Diseases* online. Consisting of data provided by the authors to benefit the reader, the posted materials are not copyedited and are the sole responsibility of the authors, so questions or comments should be addressed to the corresponding author.

Notes

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Potential conflicts of interest. All authors: No reported conflicts of interest. All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

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