Taylor & Francis
Taylor & Francis Group

Check for updates

ORIGINAL ARTICLE

Characteristics and laboratory findings on admission to the emergency department among 2873 hospitalized patients with COVID-19: the impact of adjusted laboratory tests in multicenter studies. A multicenter study in Spain (BIOCOVID-Spain study)

Luis García de Guadiana-Romualdo[a], Daniel Morell-García[b], Cristian Morales-Indiano[c], Josep Miquel Bauça[b], María José Alcaide Martín[d], Clara Esparza del Valle[e], Jose I. Gutiérrez Revilla[f], Eloisa Urrechaga[g], José M. Álamo[h], Ana M. Hernando Holgado[i], María-Carmen Lorenzo-Lozano[j], Silvia Sánchez Fdez-Pacheco[k], Patricia de la Hera Cagigal[l], María A. Juncos Tobarra[m], Juan A. Vílchez[n], Isabel Vírseda Chamorro[o], Irene Gutiérrez Garcia[p], Yolanda Pastor Murcia[q], Laura Sahuquillo Frías[r], Laura Altimira Queral[s], Elisa Nuez-Zaragoza[t], Juan Adell Ruiz de León[u], Alicia Ruiz Ripa[v], Paloma Salas Gómez-Pablos[w], Iria Cebreiros López[x], Amaia Fernández Uriarte[y], Álex Larruzea[z], María L. López Yepes[aa], Patricia Esteban Torrella[ab], María C. Zamorano Andrés[ac], Jose Pedregosa Díaz[ad], Cristina Acevedo Alcaraz[ae], Alfonso-L. Blazquez-Manzanera[af], Ana M. L. Padilla Jiménez[e], María C. Baamonde Calzada[f], Marina Vera[h], Matilde Cháfer Rudilla[j], Magdalena Canalda Campás[k], Sara García Muñoz[l], Luis Vicente Gutiérrez[m], Laura Jiménez Añón[c], Alfonso Pérez Martínez[n], Aurelio Pons Castillo[o], Ruth González Tamayo[p], Jorge Férriz Vivancos[q], Olaia Rodríguez-Fraga[d], Vicente Ferrer Díaz de Brito Fernández[s], Vicente Aguadero[t], María G. García Arévalo[u], María Arnaldos Carrillo[x], Mercedes González Morales[a], María Núñez Gárate[y], Cristina Ruiz Iruela[z], Natalia Sancho-Rodríguez[ab], Marti Vila Pérez[ad], José M. Egea-Caparrós[ae], Luis Sáenz[af], Álvaro Blasco Barbero[ag] and Amparo Galán Ortega[ah]

[a]Laboratory Medicine Department, Hospital Universitario Santa Lucía, Cartagena, Spain; [b]Laboratory Medicine Department, Hospital Universitario Son Espases, Palma de Mallorca, Spain; [c]Laboratory Medicine Department, Hospital Universitari Germans Trias i Pujol, Badalona, Spain; [d]Laboratory Medicine Department, Hospital Universitario La Paz, Madrid, Spain; [e]Laboratory Medicine Department, Hospital Universitario Marqués de Valdecilla, Santander, Spain; [f]Department of Clinical Biochemistry, Hospital Sierrallana, Torrelavega, Spain; [g]Biocruces Bizkaia Health Research Institute, Baracaldo, Spain; [h]Biochemical Laboratory, Hospital Marina Baixa, Villajoyosa, Spain; [i]Laboratory Medicine Department, Hospital Universitario Vinalopó, Elche, Spain; [j]Laboratory Medicine Department, Complejo Hospitalario de Toledo, Toledo, Spain; [k]Laboratory Medicine Department, Hospital Central de la Defensa Gómez Ulla, Madrid, Spain; [l]Laboratory Medicine Department, Hospital Universitario de Basurto, Bilbao, Spain; [m]Laboratory Medicine Department, Complejo Hospitalario Universitario de Albacete, Albacete, Spain; [n]Laboratory Medicine Department. Hospital, Universitario Morales Meseguer, Murcia, Spain; [o]Laboratory Medicine Department, Hospital Arnau de Vilanova, Valencia, Spain; [p]Laboratory Medicine Department, Hospital Universitario de Torrevieja, Torrevieja, Spain; [q]Laboratory Medicine Department, Consorci Hospital General Universitari de València, Valencia, Spain; [r]Laboratory Medicine Department, Hospital Can Misses, Ibiza, Spain; [s]Laboratory Medicine Department, Parc Sanitari Sant Joan de Déu, Sant Boi de Llobregat, Spain; [t]Clinical Laboratory Department, Parc Tauli Hospital Universitari, Sabadell, Spain; [u]Laboratory Medicine Department, Hospital Universitario Virgen de la Victoria, Málaga, Spain; [v]Laboratory Medicine Department, Hospital de Mataró, Mataró, Spain; [w]Catlab. Consorci Sanitari de Terrassa, Terrassa, Spain; [x]Laboratory Medicine Department, Hospital Universitario Virgen de la Arrixaca, Murcia, Spain; [y]Catlab. Hospital Universitario Mútua Terrassa, Barcelona, Spain; [z]Laboratory Medicine Deparment, Hospital Fundació Sanitària Mollet, Barcelona, Spain; [aa]Laboratory Medicine Department, Hospital Virgen del Castillo, Yecla, Spain; [ab]Laboratory Medicine Department. Hospital General, Universitario Reina Sofía, Murcia, Spain; [ac]Laboratory Medicine Department, Hospital de la Vega Lorenzo Guirao, Cieza, Spain; [ad]Laboratory Medicine Department, Hospital Verge de la Cinta, Tortosa, Spain; [ae]Laboratory Medicine Department. Hospital, Universitario Los Arcos del Mar Menor, San Javier, Spain; [af]Laboratory Medicine Department. Hospital General, Universitario Rafael Méndez, Lorca, Spain; [ag]Clinical Laboratory, Hospital Universitari Sant Joan, Alicante, Spain; [ah]Comisión de Magnitudes Biológicas relacionadas con la Urgencia Médica, Sociedad Española de Medicina de Laboratorio (SEQC-ML), Barcelona, Spain

ABSTRACT
Identification of predictors for severe disease progression is key for risk stratification in COVID-19 patients. We aimed to describe the main characteristics and identify the early predictors for severe outcomes among hospitalized patients with COVID-19 in Spain. This was an observational, retrospective cohort study (BIOCOVID-Spain study) including COVID-19 patients admitted to 32 Spanish hospitals. Demographics, comorbidities and laboratory tests were collected. Outcome was in-hospital mortality. For analysis, laboratory tests values were previously adjusted to assure the comparability of results among participants. Cox regression was performed to identify predictors. Study population included 2873 hospitalized COVID-19 patients. Nine variables were independent predictors for in-hospital mortality, including creatinine (Hazard ratio [HR]:1.327; 95% Confidence Interval [CI]: 1.040-1.695,

CONTACT Luis García de Guadiana-Romualdo ✉ guadianarom@yahoo.es 📍 Laboratory Medicine Department, Hospital Universitario Santa Lucía, C/Mezquita, s/n, Paraje Los Arcos, Santa Lucía, 30202, Cartagena

Supplemental data for this article can be accessed here.

$p = .023$), troponin (HR: 2.150; 95% CI: 1.155-4.001; $p = .016$), platelet count (HR: 0.994; 95% CI: 0.989-0.998; $p = .004$) and C-reactive protein (HR: 1.037; 95% CI: 1.006-1.068; $p = .019$). This is the first multi-center study in which an effort was carried out to adjust the results of laboratory tests measured with different methodologies to guarantee their comparability. We reported a comprehensive information about characteristics in a large cohort of hospitalized COVID-19 patients, focusing on the analytical features. Our findings may help to identify patients early at a higher risk for an adverse outcome.

Introduction

In December 2019, severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) was identified as the etiological agent for the pneumonia cases of unknown origin in Wuhan (Hubei Province, China), a disease designated as coronavirus disease 2019 (COVID-19) [1]. On March 11th, COVID-19 was characterized as a pandemic. Spain remains one of the European countries most severely affected by the ongoing COVID-19 pandemic both in terms of number of infected individuals and in terms of number of deaths [2].

The clinical presentation of COVID-19 varies from asymptomatic or mild upper respiratory tract symptoms to severe viral pneumonia with respiratory failure and death. The real challenge for the clinicians in an Emergency Department (ED) is to early identify COVID-19 patients at high risk for an unfavorable progression and to ensure optimal resource allocation. Therefore, there is a need to identify risk factors for early prediction of progression of COVID-19 patients.

The role of clinical laboratories in this viral outbreak includes staging, prognostication and therapeutic monitoring of individuals with COVID-19. A recent systematic review highlighted the most important biochemical and hematological alterations observed in these patients, with great interest for prognostic purposes [3]. In this line, different laboratory tests have been identified as outcome-related predictors, useful to support medical decision making [4,5]. Nonetheless, most studies referenced suffer from limitations, including small sample sizes, study population selection bias, great heterogeneity across the studies included in the reviews and meta-analysis and use of different endpoints of severity [5]. Besides, most of conclusions to date have been reported from studies performed in Chinese populations and variations in rates for COVID-19 hospitalizations and mortality across different locations suggesting differences in population characteristics have previously been reported [6].

In addition, much of the published investigation does not include information about analytical methods used for testing [5]. Hence, the International Federation of Clinical Chemistry and Laboratory Medicine (IFCC) Task Force in COVID-19 has recently urged caution when translating study findings to local laboratory practice. Of note, in multi-center studies, the use of a wide variety of methodologies and analyzers for measurement of biomarkers implies a high data heterogeneity and a poor reproducibility and only the use of adjusted results will enable the universal application of decision limits obtained from different methodologies undertaken in different locations [7,8].

BIOCOVID-Spain study is an initiative by Laboratory Medicine professionals in Spain to generate a multicenter cohort database focusing on laboratory tests. From this information, we aimed to describe the characteristics and main laboratory findings of hospitalized COVID-19 patients in Spain and identify early clinical and laboratory predictors of in-hospital mortality.

Material and methods

Study design

BIOCOVID-Spain study is a multicenter, retrospective observational study including patients hospitalized with a diagnosis of COVID-19 recruited in 32 hospitals of Health National System in 9 autonomous communities of Spain. The recruitment period was from 1 March 2020, to April 30, 2020. The follow-up censoring date was 20 May 2020. The study was approved by the Ethics Committee of all participating hospitals. Because of the retrospective design, we received the approval for data collection with waiver of informed consent.

This study was endorsed by Spanish Association of Medical Biopathology and Laboratory Medicine (AEBM-ML), Spanish Association of Clinical Laboratory (AEFA) and Spanish Society of Laboratory Medicine (SEQC-ML).

Study population

All consecutive adult patients (≥ 14 years) discharged or dead after hospital admission, with SARS-CoV-2 infection, were eligible for inclusion in the study. COVID-19 was diagnosed either by positive result of real-time reverse transcriptase-polymerase chain reaction (RT-PCR) testing of a nasopharyngeal specimen, or by a positive result of serological testing and a clinically compatible presentation.

Exclusion criteria were: (a) patients <14 years; (b) pregnant women; (c) patients transferred from or to another hospital; (d) patients transferred from nursing homes; (e) patients discharged from the ED for at home treatment; and (f) patients with Intensive Care Unit (ICU) admission criteria who were not admitted due to lack of availability.

Data collection

Data collection was performed retrospectively from electronic medical records and laboratory information systems by two researchers for each hospital. For eligible patients, we extracted the demographic information, comorbidities, laboratory test results, and disposition (discharge to home and in-hospital mortality). Laboratory data depicted the first test result occurring within the first 24 h from presentation

to the ED. A multicenter database was prepared for register of collected data and all patient identities were coded for blindness.

Outcome measures

The outcome was all-cause in-hospital mortality occurring during follow-up.

Adjustment of laboratory tests

To achieve an adjustment of laboratory test values, a harmonization factor was calculated for each methodology and analyzer, using a category 1 external quality assurance (EQA) scheme, provided by the Spanish Society of Laboratory Medicine (SEQC-ML), which includes results that are interchangeable and traceable to reference standards [9]. The value measured with the most common methodology and analyzer among participating laboratories was considered as reference to generate the factor. For those biochemical tests not included in this quality control material (albumin, C-reactive protein [CRP], ferritin, urea and lactate), a correction factor was calculated from other nonconmutable EQA schemes of the SEQC-ML (See Supplementary Material). To assure the comparability in a wide measurement range, a mean factor was calculated from the factors calculated in 6 EQA materials send to laboratories during 2019. A similar approximation has been recently performed by Kurstjens et al. [10] in a study evaluating the role of biochemical and hematological tests for a rapid identification of SARS-CoV-2-infected patients, in which significant differences were observed for ferritin measurement when different analyzers were used.

For cardiac troponin, only high sensitivity assays were included and the levels were dichotomized according to sex-stratified 99th-percentile. For procalcitonin, only Brahms-like methods were included.

For D-dimer, according to Favaloro et al., results were converted to a single unit of measurement (ng/mL Fibrinogen Equivalent Units [FEU]) [11]. Adjustment of all other hematological and coagulation tests was considered as not necessary according to the EQA scheme provided by the Spanish Society of Hematology and Hemotherapy (SEHH).

Statistical analysis

Continuous variables were tested for normal distribution using the Kolmogorov-Smirnov's test. Data are summarized as numbers and frequencies for categorical variables and or medians with interquartile ranges (IQRs) for continuous data. Comparisons between groups were performed with Chi-squared test for categorical data and Mann-Whitney U tests for continuous data. Cox regression analysis was performed to identify variables associated with in-hospital mortality. To obtain the predictors, we performed a block-wise forward procedure allocating the predictor variables into three clusters: demographics, comorbidities and laboratory tests. A multivariable regression analysis was fitted within each block using statistical significance (p-value <.10) as

criterion to achieve the best set of predictors. Statistical significance was set at 5%. SPSS software version 20 (IBM Corporation, USA) was used for all statistical analyses.

Results

Characteristics of study subjects

During the study period, a total of 2981 COVID-19 patients admitted to 32 Spanish hospitals were recruited. One-hundred and eight patients who were still hospitalized on May 20, 2020 were excluded from analyses. The study population finally included 2873 hospitalized COVID-19 patients.

In-hospital mortality rate was 14.3%, which was seen to be significantly higher in male compared to female sex (16.4% vs. 11.3%; $p < .001$). ICU admission was required for 504 patients (17.5%); in these patients in-hospital mortality rate was 35.1%, significantly higher than in non-admitted to ICU (9,9%; $p < .001$). The median time from ED admission to ICU admission was 2 days (IQR: 1-4), while in-hospital length of stay and ICU length of stay were 22 days (IQR: 15-34) and 12 days (IQR: 6-20), respectively.

Patients' characteristics, stratified by previously defined outcomes, are summarized in Table 1. Median age was 66 years (IQR: 54–76), ranging from 15 to 98 years and 41.7% were ≥70 years; 1699 patients were male (59.1%). The most common comorbidity was hypertension (45.8%), followed by diabetes mellitus (24.3%) and cardiovascular disease (CVD) (22.6%) and 1722 (59.9%) patients presented at least one comorbidity.

Patients who died were older and with a higher frequency of male sex and all comorbidities; 339 (82.3%) had at least a comorbidity. In-hospital mortality increased with age, reaching a percentage of 29.9% for patients over 80 years.

Laboratory findings

Individuals with altered baseline laboratory tests are presented in Table 2, also stratified according to the outcomes. The most common laboratory alterations were increased

Table 1. Demograhics and comorbidities of 2873 COVID-19 patients grouped according to in-hospital mortality at study censoring date.

	Total population n = 2873	Survivors n = 2461 (85.7%)	Non-survivors n = 412 (14.3%)	p Value
Age, years [Median (IQR)]				<.001
Distribution	66 (54–76)	63 (52–74)	76 (68–83)	<.001
<30 years	45 (1.6%)	44 (1.8)	1 (0.2)	
30–39 years	131 (4.6)	128 (5.2)	3 (0.7)	
40–49 years	343 (11.9)	338 (13.7)	5 (1.2)	
50–59 years	511 (17.8)	484 (19.7)	27 (6.6)	
60–69 years	674 (23.5)	593 (24.1)	81 (19.7)	
70–79 years	678 (23.6)	530 (21.5)	148 (35.9)	
≥80 years	491 (17.1)	344 (14.0)	147 (35.7)	
Sex, male [n (%)]	1699 (59.1)	1420 (57.7%)	279 (67.7%)	<.001
Hypertension [n (%)]	1317 (45.8)	1043 (42.4%)	274 (66.5%)	<.001
Diabetes mellitus [n (%)]	697 (24.3)	548 (22.3%)	149 (36.2%)	<.001
COPD [n (%)]	247 (8.6)	182 (7,4%)	65 (15,8%)	<.001
CVD [n (%)]	649 (22.6)	485 (19.7%)	164 (39.8%)	<.001
CKD [n (%)]	247 (8.6)	161 (6,5%)	86 (20,9%)	<.001

COPD: chronic obstructive pulmonary disease; CVD: cardiovascular disease; CKD: chronic kidney disease.

Table 2. Frequency of altered laboratory tests, measured on hospital admission, in 2873 COVID-19 patients grouped according to in-hospital mortality at study censoring date.

Variable	Total population N = 2873		Survivors n = 2461 (85.7%)		Non-survivors n = 412 (14.3%)		
	n	Number (%)	n	Number (%)	n	Number (%)	p Value
Lactate ≥ 2.0 mmol/L	1948	494 (25.4)	1670	372 (22.3)	278	122 (43.9)	<.001
Urea > 8.16 mmol/L	2748	744 (27.1)	2356	520 (22.1)	392	224 (57.1)	<.001
Creatinine, μmol/L							
>68.63 (female)	2859	799 (27.9)	2450	583 (23.8)	409	215 (52.6)	<.001
>91.51 (male)							
Albumin < 35 g/L	1366	424 (31.0)	1167	323 (27.7)	199	101 (50.8)	<.001
ALT/GPT, U/L							
>41 (male)	2687	927 (34.5)	2305	806 (35)	382	121 (31.7)	.210
>33 (female)							
AST/GOT, U/L							
>40 (male)	2068	1012 (48.9)	1771	829 (46.8)	297	183 (61.6)	.001
>32 (female)							
Bilirubin > 20.52 μmol/L	2253	109 (4.8)	1934	86 (4.4)	319	23 (7.2)	.033
CK, U/L							
>190 (male)	1644	354 (21.5)	1406	273 (19.4)	238	81 (34)	<.001
>170 (female)							
LDH, U/L							
>241 (male)	2477	1905 (76.9)	2122	1591 (75)	355	314 (88.5)	<.001
>225 (female)							
Troponin ≥ 99th percentile (sex-stratified)	1280	400 (31.3)	1078	280 (26.0)	202	120 (59.4)	<.001
CRP > 5.0 mg/L	2788	2671 (95.8)	2386	2273 (95.3)	402	398 (99)	.001
Procalcitonin ≥ 0.25 μg/L[a]	2120	499 (23.5)	1808	332 (18.4)	312	167 (53.5)	<.001
Ferritin	1806						
>500 μg/L[b]		1096 (60.7)	1581	927 (58.6)	225	169 (75.1)	<.001
>2000 μg/L[c]		241 (13.3)		197 (12.5)		44 (19.6)	.003
Hemoglobin, g/L							
<130 (male)	2869	559 (19.5)	2457	413 (16.8)	412	146 (35.4)	<.001
<120 (female)							
WBC count > 11.00 * 10⁹/L	2870	373 (13)	2459	270 (11.0)	411	103 (25.1)	<.001
Neutrophil count > 7.50 * 10⁹/L	2870	642 (22.4)	2459	470 (19.1)	411	172 (41.8)	<.001
Lymphocyte count < 1.00* 10⁹/L	2870	1430 (49.8)	2459	1155 (47.0)	411	275 (66.9)	<.001
NLR							
≥5.00 (Median)	2870	1439 (50.1)	2459	1139 (46.3)	411	300 (73)	<.001
≥8.52 (Quartile 4)		720 (25.1)		516 (21.0)		204 (49.6)	<.001
Platelet count < 150 * 10⁹/L	2870	691 (24.1)	2458	560 (22.8)	412	131 (31.8)	<.001
INR ≥1.2	2646	899 (34.0)	2265	748 (33.0)	381	151 (39.6)	.12
D-dimer > 500 μg/L FEU[d]	2663	1460 (54.8)	2299	1213 (52.8)	364	247 (67.9)	<.001

[a]Ref. [12]; [b]Ref. [13]; [c]Ref. [14].
[d]For patients ≥50 years, threshold was defined according to the formula proposed by Douma et al..
ALT/GPT: alanine aminotransferase; AST/GOT: aspartate aminotransferase; CK: creatine kinase; CRP: C-reactive protein; FEU: Fibrinogen Equivalent Units; INR, International normalized ratio; LDH, lactate dehydrogenase; NLR: neutrophil-to-lymphocyte ratio; WBC: white blood cell.

values of CRP (95.8%), lactate dehydrogenase [LDH] (76.9%), ferritin (60.7%), D-dimer (54.8%), neutrophil to lymphocyte ratio [NLR] (50.1%) and aspartate aminotransferase [AST/GOT] (48.9%), as well as a decreased lymphocyte count (49.8%).

Table 3 shows the laboratory findings upon admission in the entire cohort and stratified by survival. In non-survivor patients, lactate, urea, creatinine, AST/GOT, LDH, CRP, procalcitonin, ferritin, International Normalized Ratio [INR], D-dimer levels, as well as white blood cell (WBC) and neutrophil count, NLR and cardiac troponin levels above 99th percentile, were significantly higher compared with survivors. In contrast, albumin levels and lymphocyte and platelet counts were significantly lower. No significant differences were observed for alanine aminotransferase [ALT/GPT] and bilirubin.

Predictors of in-hospital mortality

In univariate analysis, the following variables showed a significant hazard ratio (HR) for in-hospital mortality and

were included in the multivariate analysis: stratified-age, male sex, hypertension, diabetes mellitus, chronic obstructive pulmonary disease (COPD), CVD, chronic kidney disease (CKD), urea, creatinine, albumin, AST/GOT, bilirubin, creatine kinase (CK), LDH, troponin, CRP, procalcitonin, D-dimer, ferritin and hemoglobin levels, NLR, INR and white blood cell, neutrophil, lymphocyte and platelet counts. Independent predictors for in-hospital mortality are presented in Table 4. In the final adjusted analysis by Cox regression analysis, we found 9 variables independently associated with an increased hazard of in-hospital mortality: older age, male sex, hypertension, CVD, CKD, creatinine, CRP and troponin levels and platelet count.

Discussion

Alterations in laboratory tests can be used in clinical practice to predict the severity of COVID-19 and could improve prognosis and decrease the mortality rates, but further worldwide research is needed to better understand these changes and their relations with prognosis [15]. In this

Table 3. Laboratory findings on admission of 2873 COVID-19 patients grouped according to in-hospital mortality at study censoring date.

Variable, median (IQR)	Total population n = 2873	Survivors n = 2461 (85.7%)	Non-survivors n = 412 (14.3%)	p Value
Lactate (mmol/L)	1.51 (1.10–1.98)	1.40 (1.08–1.89)	1.80 (1.30–2.50)	<.001
Urea (mmol/L)	5.99 (4.33–8.33)	3.99 (4.33–7.66)	8.66 (6.49–13.49)	<.001
Creatinine (μmol/L)	79.56 (64.53–101.66)	77.79 (63.65–96.36)	102.54 (78.68–138.79)	<.001
Albumin (g/L)	38 (34–41)	38 (35–42)	35 (31–39)	<.001
ALT/GPT (U/L)	28 (18–46)	28 (18–46)	26 (17–45)	.162
AST/GOT (U/L)	35 (25–53)	34 (24–51)	43 (29–64)	<.001
Bilirubin (μmol/L)	8.38 (5.99–12.65)	8.38 (5.99–12.65)	8.89 (6.33–13.34)	.139
CK (U/L)	88 (53–163)	85 (51–150)	116 (65–247)	<.001
LDH (U/L)	306 (237–408)	295 (232–387)	392 (290–555)	<.001
Troponin ≥99th percentile (sex-stratified)[a]	400 (31.3)	280 (26)	120 (59.4)	<.001
CRP (mg/L)	77.8 (33.7–147.9)	68.7 (30.0–135.8)	136.4 (75.1–205.9)	<.001
Procalcitonin (μg/L)	0.10 (0.06–0.23)	0.10 (0.05–0.19)	0.10 (0.11–0.68)	<.001
Ferritin (μg/L)	674 (325–1368)	647 (310–1306)	924 (500–1815)	<.001
Hemoglobin (g/L)	139 (127–150)	14 (129–150)	135 (119–147)	<.001
WBC count (* 10 9/L)	6.60 (4.94–9.00)	6.50 (4.90–8.63)	7.90 (5.50–11.04)	<.001
Neutrophil count (* 10 9/L)	4.90 (3.41–7.10)	4.72 (3.32–6.75)	6.56 (4.17–9.53)	<.001
Lymphocyte count (* 10 9/L)	1.00 (0.70–1.33)	1.00 (0.70–1.39)	0.77 (0.51–1.10)	<.001
NLR	5.0 (3.06–8.52)	4.66 (2.89–7.76)	8.43 (4.80–14.65)	<.001
Platelet count (* 10 9/L)	195 (152–256)	197 (154–259)	187 (137–245)	<.001
INR	1.12 (1.03–1.23)	1.12 (1.03–1.23)	1.13 (1.04–1.28)	.010
D-dimer (μg/L FEU)	700 (436–1241)	660 (422–1127)	1118 (582–2319)	<.001

[a]Expressed as n (%).
ALT/GPT: alanine aminotransferase; AST/GOT: aspartate aminotransferase; CK: creatine kinase; CRP: C-reactive protein; FEU: Fibrinogen Equivalent Units; INR, International normalized ratio; LDH, lactate dehydrogenase; NLR, neutrophil-to-lymphocyte ratio; WBC, white blood cell.

Table 4. Independent predictors for in-hospital mortality.

	Multivariate HR	CI 95%	p Value
Demographics			
Sex, male	1.391	1.128–1.714	.002
Age (ref. group: <50 years)			
50–64 years	3.441	1.712–6.914	.001
65–79 years	8.370	4.286–16.348	<.001
≥80 years	15.495	7.894–30.416	<.001
Comorbidities			
Hypertension	1.582	1.272–1.967	<.001
CVD	1.658	1.343–2.047	<.001
CKD	1.898	1.477–2.440	<.001
Laboratory tests			
Creatinine	1.327	1.040–1.695	.023
Troponin ≥ 99th percentile	2.150	1.155–4.001	.016
CRP	1.037	1.006–1.068	.019
Platelet count	0.994	0.989–0.998	.004

CVD: Cardiovascular disease; CKD: Chronic kidney disease; CRP: C-reactive protein; HR: Hazard ratio; CI: Confidence interval.

study, we describe the characteristics of 2873 hospitalized patients by SARS-CoV-2 infection from different regions of Spain, focusing on the alterations in laboratory tests.

In our study, in-hospital mortality was 14.3%; whereas the reported mortality rate in other multicenter Spanish populations for hospitalized patients has ranged from 11.9 to 28% [16–18]. This variability is also observed for international cohorts of hospitalized COVID-19 patients, with mortality rates varying from 1.4% in Chinese series [19] to 17% [20] and 26% [21] in Western populations. Differences in mortality rate reported have been attributed to causes as the different demographic characteristics and prevalence of comorbidities, the differing strategies used for SARS-CoV-2 RT-PCR testing and how COVID-19-related deaths were identified [22]. However, differences between Spanish series only could be explained by different criteria for inclusion of patients in the studies and for admission to hospitals, according to differences in the availability of health resources.

Concerning to demographics, age in our cohort was similar to that reported in other national studies [16–18], although lower than that found by Docherty et al. [21] in United Kingdom, with a median age of 73 years. Of note, Chinese studies have reported even clearly lower ages, below 50 years [19]. Similar to the findings recently highlighted by Bonanad et al. [23], an increase of age-related mortality, with the highest mortality rate among patients older than 80 years, was described in this cohort.

Male sex has been also reported as a risk factor for severity of COVID-19 [24]. As described in previous studies [16–18,21], the number of male patients was higher than female and death was significantly higher in men. It would be related with sex differences in immune response that might impact on the inflammatory response and outcomes of COVID-19 [25].

Pre-existing conditions are described as significant predictors of COVID-19 disease outcomes in the literature [26]. Similar to previous findings described in Western and Asian cohorts [16–21], the main comorbidities in our study were a history of hypertension and diabetes mellitus. Unfortunately, other common comorbidities in COVID-19, such as dyslipidemia or obesity, were not collected in this study. Besides, a high proportion of patients, almost 60%, had at least one comorbidity, although this proportion was lower than that reported recently by Berenguer et al. [16], reaching a rate of 73.8%, likely due to the inclusion of a greater number of comorbidities. Similar to previous studies, comorbidities such as hypertension, CVD and CKD were predictors of in-hospital mortality [16]; however, of note, diabetes mellitus was not a predictor for this outcome, as Pugliese et al. recently suggested [27].

Concerning to laboratory tests, the clinical laboratories involved in this study used different analyzers from the major in vitro diagnostic device suppliers. Although several multicenter studies have reported the main blood

biochemical, hematologic and coagulation features of COVID-19, no prior adjustment of the results was performed to ensure the comparability of the results between the participating laboratories [16–19]. This lack of adjustment can generate data misinterpretation and compromise the robustness of conclusions reported [28]. Laboratory tests such LDH [7], ferritin [10] and albumin [29], reported as predictors of severity in COVID-19 patients, are strongly affected by the analytical method used to measure its blood levels. Therefore, to apply an adjustment factor contributes to the comparability of results.

We describe the presence of altered values for laboratory tests since the initial stage of early infection was a common finding. The most frequent alterations were increased values of CRP, LDH, ferritin and D-dimer, as well as a decreased lymphocyte count, considered as a signature for severe COVID-19 [30]. In our study, the rate of patients with abnormalities in these inflammatory markers was higher in patients who died. This finding could be related to the cytokine storm, already known in patients with severe to critical disease, in which lymphocyte count sharply decreased alongside with elevations of D-dimer, CRP and ferritin [31]. A high number of patients also had alterations in other laboratory tests since the early stages of infection, with abnormalities of renal, hepatic, cardiac or coagulation markers. Similar findings were found for procalcitonin, in agreement with previous studies [17], whose circulating levels should be in normal range, as expected for a viral infection, and lactate levels, for which a potential role for prognosis has hardly been analyzed so far.

The significant difference detected for most of laboratory tests levels in patients with a poor outcome is a consistent finding with other reports [16,17,21] and suggest a potential role for prognosis of COVID-19 patients. We identified several routine laboratory markers as predictors of in-hospital mortality, such as creatinine, troponin, CRP and platelet count, which have been suggested as predictors in previous studies and reviews [32–35]. We would like to highlight the role of creatinine as predictor for in-hospital mortality, suggested in previous studies [36]. Acute or pre-existing renal disease have been previously reported as predictors for the development of severe COVID-19 [36]. Although the kidney injury in COVID-19 is multifactorial, SARS-CoV-2 using angiotensin converting enzyme 2 (ACE2) receptor for renal cell entry and the virus-induced cytokines causing indirect effects on renal cells *via* shock or hypoxia have been proposed as mechanisms to explain the acute renal injury in these patients [37].

Strenghts of this study, in addition to the sample size, are the previous estimation of this and the effort to adjust the results to guarantee their comparability. To our knowledge, only Kurstjens et al., performed a similar approach, calculating a harmonization factor to correct the observed differences for ferritin levels measured in different analyzers [10]. Besides, laboratory test levels were stratified by sex and age, as appropriate and recommended for tests such as D-dimer [38].

Our study has a number of limitations. First, this is a retrospective study and subject to the limitations of this design, including selection bias, errors in data entry, and residual confounding. Second, during the COVID-19 outbreak, a standardized analytical profile for ED or admission to ward or ICU was not establshed at onset or during the pandemic in all the participating centers and the available laboratory tests for physicians varied widely between hospitals; therefore, some cases had incomplete documentation of laboratory testing and the number of available results for each laboratory test was variable. Third, the role of IL-6, previously reported as predictor of COVID-19 severity [39], was not evaluated in our study, because it was measured in a small number of patients and with a great methodological variability.

Conclusion

In summary, analysis of laboratory data in multicenter studies requires a previous adjustment of results to assure the comparability of results reported by different laboratories. In our cohort, including a large number of hospitalized COVID-19 patients, we identified early risk factors for a poor outcome, including laboratory tests available in most of laboratories. These findings could help in the proper hospital management from the ED in patients with SARS-CoV-2 infection to early identify those at a high risk for severe disease progression.

Author contributions

LGGR conceived and designed the study and supervised the conduct of the trial and data collection. DMG and ABB provided statistical advice on study design and analyzed the data. LGGR and DMG drafted the article, and CMI, JMBR and MJAM contributed substantially to its revision. All the authors contributed to data collection in the participating hospitals and final approval of this article.

Disclosure statement

All authors report no potential conflict of interest. This research did not receive any specific grant from funding agencies in the public, comercial, or not-for-profits sectors

References

[1] Zhu N, Zhang D, Wang W, et al.; China Novel Coronavirus Investigating and Research Team. A novel coronavirus from patients with pneumonia in China, 2019. N Engl J Med. 2020; 382(8):727–733.

[2] Instituto de Salud Carlos III. COVID-19 in Spain. 2020. https://cnecovid.isciii.es/covid19

[3] Kavsak PA, de Wit K, Worster A. Emerging key laboratory tests for patients with COVID-19. Clin Biochem. 2020;81: 13–14.

[4] Zhang ZL, Hou YL, Li DT, et al. Laboratory findings of COVID-19: a systematic review and meta-analysis. Scand J Clin Lab Invest. 2020;80(6):441–447.

[5] Thompson S, Bohn MK, Mancini N, et al.; IFCC Taskforce on COVID-19. IFCC interim guidelines on biochemical/hematological monitoring of COVID-19 patients. Clin Chem Lab Med. 2020;58(12):2009–2016.

[6] Wadhera RK, Wadhera P, Gaba P, et al. Variation in COVID-19 hospitalizations and deaths across New York City Boroughs. Jama. 2020;323(21):2192–2195.

[7] Panteghini M. Lactate dehydrogenase: an old enzyme reborn as a COVID-19 marker (and not only). Clin Chem Lab Med. 2020;58(12):1979–1981.

[8] van der Burg SH, Kalos M, Gouttefangeas C, et al. Harmonization of immune biomarker assays for clinical studies. Sci Transl Med. 2011;3(108):108ps44–108ps44.

[9] Ricós C, Fernández-Calle P, Marqués F, et al. Impact of implementing a category 1 external quality assurance scheme for monitoring harmonization of clinical laboratories in Spain. Advances in Laboratory Medicine/Avances en Medicina de Laboratorio. 2020;1:20200008.

[10] Kurstjens S, van der Horst A, Herpers R, et al. Rapid identification of SARS-CoV-2-infected patients at the emergency department using routine testing. Clin Chem Lab Med. 2020;58(9):1587–1593.

[11] Favaloro EJ, Thachil J. Reporting of D-dimer data in COVID-19: some confusion and potential for misinformation. Clin Chem Lab Med. 2020;58(8):1191–1199.

[12] Neeser O, Branche A, Mueller B, et al. How to: implement procalcitonin testing in my practice. Clin Microbiol Infect. 2019;25(10):1226–1230.

[13] Henter JI, Horne A, Aricó M, et al. HLH-2004: diagnostic and therapeutic guidelines for hemophagocytic lymphohistiocytosis. Pediatr Blood Cancer. 2007;48(2):124–131.

[14] Sarangi R, Pathak M, Padhi S, et al. Ferritin in hemophagocytic lymphohistiocytosis (HLH): current concepts and controversies. Clin Chim Acta. 2020;510:408–415.

[15] Kermali M, Khalsa RK, Pillai K, et al. The role of biomarkers in diagnosis of COVID-19 - A systematic review . Life Sci. 2020;254:117788.

[16] Berenguer J, Ryan P, Rodríguez-Baño J, et al.; Centro Nacional de Epidemiología. Characteristics and predictors of death among 4035 consecutively hospitalized patients with COVID-19 in Spain . Clin Microbiol Infect. 2020;26(11):1525–1536.

[17] Gil-Rodrigo A, Miró Ò, Piñera P, et al. en representación de la red de investigación SIESTA. Analysis of clinical characteristics and outcomes in patients with COVID-19 based on a series of 1000 patients treated in Spanish emergency departments. Emergencias. 2020;32:233–241.

[18] Casas-Rojo JM, Antón-Santos JM, Millán-Núñez-Cortés J, et al.; en nombre del Grupo SEMI-COVID-19 Network. Clinical characteristics of patients hospitalized with COVID-19 in Spain: results from the SEMI-COVID-19 Registry. Rev Clin Esp. 2020;220(8):480–494.

[19] Guan WJ, Ni ZY, Hu Y, et al. China medical treatment expert group for covid-19. Clinical characteristics of coronavirus disease 2019 in China. N Engl J Med. 2020;382(18):1708–1720.

[20] Kim L, Garg S, O'Halloran A, et al. Risk factors for intensive care unit admission and In-hospital mortality among hospitalized adults identified through the U.S. coronavirus disease 2019 (COVID-19)-associated hospitalization surveillance network (COVID-NET). Clin Infect Dis. 2020;ciaa1012.

[21] Docherty AB, Harrison EM, Green CA, ISARIC4C investigators, et al. ISARIC4C investigators. Features of 20 133 UK patients in hospital with covid-19 using the ISARIC WHO Clinical Characterisation Protocol: prospective observational cohort study. BMJ. 2020;369:m1985.

[22] Onder G, Rezza G, Brusaferro S. Case-Fatality Rate and Characteristics of Patients Dying in Relation to COVID-19 in Italy. Jama. 2020;323(18):1775–1776.

[23] Bonanad C, García-Blas S, Tarazona-Santabalbina F, et al. The effect of age on mortality in patients with COVID-19: a meta-analysis with 611,583 subjects. J Am Med Dir Assoc. 2020;21(7):915–918.

[24] Jin JM, Bai P, He W, et al. Gender differences in patients with COVID-19: focus on severity and mortality. Front Public Health. 2020;8:152.

[25] Klein SL, Flanagan KL. Sex differences in immune responses. Nat Rev Immunol. 2016;16(10):626–638.

[26] Zhou Y, Yang Q, Chi J, et al. Comorbidities and the risk of severe or fatal outcomes associated with coronavirus disease 2019: A systematic review and meta-analysis. Int J Infect Dis. 2020;99:47–56.

[27] Pugliese G, Vitale M, Resi V, et al. Is diabetes mellitus a risk factor for Corona virus disease 19 (COVID-19)? Acta Diabetol. 2020;57(11):1275–1211.

[28] Plebani M. Harmonization in laboratory medicine: the complete picture. Clin Chem Lab Med. 2013;51(4):741–751.

[29] Aloisio E, Serafini L, Chibireva M, et al. Hypoalbuminemia and elevated D-dimer in COVID-19 patients: a call for result harmonization. Clin Chem Lab Med. 2020;58(11):e255–e256.

[30] Bermejo-Martin JF, Almansa R, Menéndez R, et al. Lymphopenic community acquired pneumonia as signature of severe COVID-19 infection. J Infect. 2020;80(5):e23–e24.

[31] Wang J, Jiang M, Chen X, et al. Cytokine storm and leukocyte changes in mild versus severe SARS-CoV-2 infection: review of 3939 COVID-19 patients in China and emerging pathogenesis and therapy concepts. J Leukoc Biol. 2020;108(1):17–41.

[32] Bahl A, Van Baalen MN, Ortiz L, et al. Early predictors of in-hospital mortality in patients with COVID-19 in a large American cohort. Intern Emerg Med. 2020;15(8):1485–1415.

[33] Gaze DC. Clinical utility of cardiac troponin measurement in COVID-19 infection. Ann Clin Biochem. 2020;57(3):202–205.

[34] Sahu BR, Kampa RK, Padhi A, et al. C-reactive protein: a promising biomarker for poor prognosis in COVID-19 infection. Clin Chim Acta. 2020;509:91–94.

[35] Lippi G, Plebani M, Henry BM. Thrombocytopenia is associated with severe coronavirus disease 2019 (COVID-19) infections: A meta-analysis. Clin Chim Acta. 2020;506:145–148.

[36] Bhargava A, Fukushima EA, Levine M, et al. Predictors for Severe COVID-19 Infection. Clin Infect Dis. 2020;71(8):1962–1968.

[37] Albini A, Di Guardo G, Noonan DM, et al. The SARS-CoV-2 receptor, ACE-2, is expressed on many different cell types: implications for ACE-inhibitor- and angiotensin II receptor blocker-based cardiovascular therapies. Intern Emerg Med. 2020;15(5):759–766.

[38] Roncon L, Zuin M, Zonzin P. Age-adjusted D-dimer cut-off levels to rule out venous thromboembolism in COVID-19 patients. Thromb Res. 2020;190:102.

[39] Laguna-Goya R, Utrero-Rico A, Talayero P, et al. IL-6-based mortality risk model for hospitalized patients with COVID-19. J Allergy Clin Immunol. 2020;146(4):799–807.e9.