SUPPLEMENTARY MATERIAL

Health care-associated infections in patients with COVID-19 pneumonia in COVID critical care areas

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Methods

Patients. We included all patients with a diagnosis of pneumonia and polymerase chain reaction (PCR) positive for COVID-19 admitted for more than 48 hours to one of the COVID-19 ICUs in our 500-bed university general hospital during the first wave of the pandemic (March 10 to May 31, 2020).

COVID-19 ICUs. This comprised four ICUs designated for the care of critically ill COVID-19 patients during the pandemic. The original multipurpose ICU had 18 individual bedrooms, two of which were occupied by two beds during the emergency period (maximum occupancy 20 beds). Each year this ICU participates in the ENVIN-HELICS registry and implements in a controlled manner (with external audits and internal safety rounds) the recommendations of the “Project Zeros” supported by the MSCBS and Spanish society of intensive and critical care units and coronary units (SEMICYUC). During the pandemic period the hospital expanded the number of beds in order to care for critically ill COVID-19 patients and repurposed three additional expanded ICUs (eICU): eICU-1, with 10 beds; eICU-2, with 19 beds; and eICU-3, with 18 beds, for a total 67 ICU beds. In some of these ICUs the boxes were smaller, some had areas that could not be isolated, and in some cases the operating room was used as a conventional box. At the start of May 2020, all patients still in the eICUs were moved to the main ICU and all the eICUs returned to their original function. All these units during the pandemic included a multidisciplinary medical team with critical care physicians, anesthetists, pulmonologists and cardiologists. The admissions were always valued and coordinated by intensivists. While they predominated in the ICU and in the eICU 1, anesthetists predominated in the eICU 2 and eICU 3.

Variables. The following information was collected from the electronic medical records: demographic details (age, gender), illness severity (APACHEII), movement during ICU stay (defined as change of location between ICUs), length of ICU stay previous to HAI (ICU LOS prior to HAI, days), total length of ICU stay (total ICU LOS, days), comorbidities, complications (including HAIs), laboratory test results, microbiology performed during ICU stay, treatments, and outcomes. *Static* patients were defined as those who were admitted to and discharged from the same ICU. *Imported* patients were those transferred to the indicated unit from another ICU and *exported* patients were those who were transferred from the indicated unit to other ICUs. Most of the recorded comorbidities included were defined according to the criteria in the ENVIN-HELICS manual (12). Other comorbid conditions were included if recorded in the patient's notes or if specific criteria were present: dyslipidaemia, if total cholesterol levels were ≥ 240 mg/dL and/or LDL-cholesterol ≥ 160 mg/dL and/or triglycerides ≥ 200 mg/dL on admission; obesity, if body mass index ≥ 30 kg/m^2; hypertension, if on treatment with antihypertensives; and hypothyroidism, if requiring hormone replacement therapy. The laboratory parameters measured in the first 24 hours of admission to the COVID-19 ICUs were C-reactive protein (CRP; mg/dL), procalcitonin (ng/mL), interleukin 6 (IL-6; pg/mL), lymphocytes (10^3 cells/µL), ferritin (ng/mL), fibrinogen (mg/dL), lactate dehydrogenase (LDH; U/L), and D-dimer (ng/mL). Treatments were classified as antivirals potentially active against COVID-19 (lopinavir-ritonavir, interferon, darunavir, and remdesivir), immunomodulators (tocilizumab, sarilumab), corticosteroids, antimicrobials, neuromuscular blockers, and vasopressors. The use of antimicrobials was quantified by days of treatment per 100 days' ICU stay (DOT). Outcomes were classified as ICU length of stay and death during ICU stay. Death was defined as related to HAI when it occurred in the 72 hours following diagnosis of the infection.

Definition of HAI. HAI was defined as infection diagnosed more than 48 hours after admission to a COVID-19 ICU and less than 48 hours after ICU discharge. HAIs were attributed to the specific COVID-19 ICU in which they were diagnosed. In patients who were moved between units, if infection was diagnosed within 48 hours of transfer, it was attributed to the unit of origin. HAIs were defined according to the ENVIN-HELICS registry manual criteria into catheter-related bloodstream infection (CRBSI), bacteraemia of unknown origin (BUO), secondary bacteraemia (SB), urinary tract infection

(UTI) and ventilator-associated lower respiratory tract infections (VLRTI) including tracheobronchitis and pneumonias (VAT and VAP) respectively (12). This study also included infection of pressure ulcers (IPU) and tracheostomy site infections (TSI). In all cases, the infections were recorded in the medical record and the microorganisms responsible were identified on blood culture or culture of biological samples from the site of infection. Patients were classified according to the presence or absence of HAI and according to the number of HAIs present during their ICU stay: 0, 1, 2, or ≥3. Microorganisms were classified as multi-drug resistant (MDR) according to the definitions in the ENVIN manual (12). In this study, isolates of *Candida parapsilopsis* that were highly-resistant to fluconazole were included as MDR.

Definition of rates. The rates of HAI are presented as follows: *Prevalence* of patients with ≥ 1 HAI, for all the COVID ICUs and for each individual ICU (percentage); *HAI incidence rate per 1000 days* (1000d), for all COVID-19 ICUs and for each ICU [(total number of HAIs / total days' stay in ICU) * 1000)]; and *incidence rate of each infection per 1000d* [(number of each HAI / total days' stay in ICU) * 1000]. In the case of VLRTI, total days of mechanical ventilation were used.

Statistical analysis. Continuous variables are expressed as mean and standard deviation (SD) or median and interquartile range (IR) depending on their parametric or non-parametric distribution. Categorical variables are reported as absolute frequencies and percentages. Differences between groups were analysed using the chi-squared or Fisher's exact test for categorical variables, and the Student t-test or Mann-Whitney U test for continuous variables, respectively. Logistic regression was used to assess independent factors associated with HAI. Statistical significance was established at a p-value <0.05. Data were analysed using the Statistical Package for Social Sciences 24.0 (IBM® SPSS Statistics®, Chicago, IL, USA) for Windows. The study was approved by the PSMAR Ethics Committee (2020/9243).

Results

Supplement online figures and tables are shown.

Discussion

The main limitations of this study are its retrospective nature, the small sample size and the difficulty in reaching a firm diagnosis of VAP, since all the patients already had bilateral pulmonary infiltrates upon admission to the ICU. To avoid underdiagnosis, we also included VAT under the category of VLRTI. This diagnosis required the presence of purulent tracheal exudate, fever, positive cultures for pathogenic microorganisms in the bronchial exudate and treatment specific for the infection. Another similar limitation was the difficulty in differentiating asymptomatic candiduria from *Candida spp* UTI; we classified it as UTI when that was the interpretation stated in the medical records and specific treatment was given. The results of this study are not comparable with the national results published in the ENVIN-HELICS registry as our study counted additional types of HAIs. Finally, as the study was performed in a single centre, the results are not generalisable to other ICUs. The data from the ENVIN-UCI registry of patients with COVID-19 should be able to confirm our findings.

Acknowledgements

We would like to thank all the health care professionals of the different critical care units for their work, unwavering dedication, and professionalism throughout (cardiologists, pulmonologists, anesthesiologists, nurses and clinical assistants as well as orderlies). To the general service staff, especially those involved in maintenance and purchasing, for their commitment and cooperation in preparing and equipping the eICU and their efforts to obtain the materials required for patient care. To the microbiology and pharmacy departments for their timely responses to the demands in relation to COVID-19 patient care, in particular Marta de Antonio (pharmacy) and Miquel Micó (microbiology) who provided data on corticosteroids and immunomodulators and the microbiological results. To Xavier Marin, Teresa Cruet, Gerard Munté and Montserrat Beltran who assisted with data collection, and Marta Gas for her administrative support and database maintenance.

Figure 1e. Flow chart of COVID-19 patients admitted to the COVID-19 intensive care units during the pandemic and included in the study.

Footnote Figure 1: PCR: polymerase chain reaction; HAI: health care-associated infection.

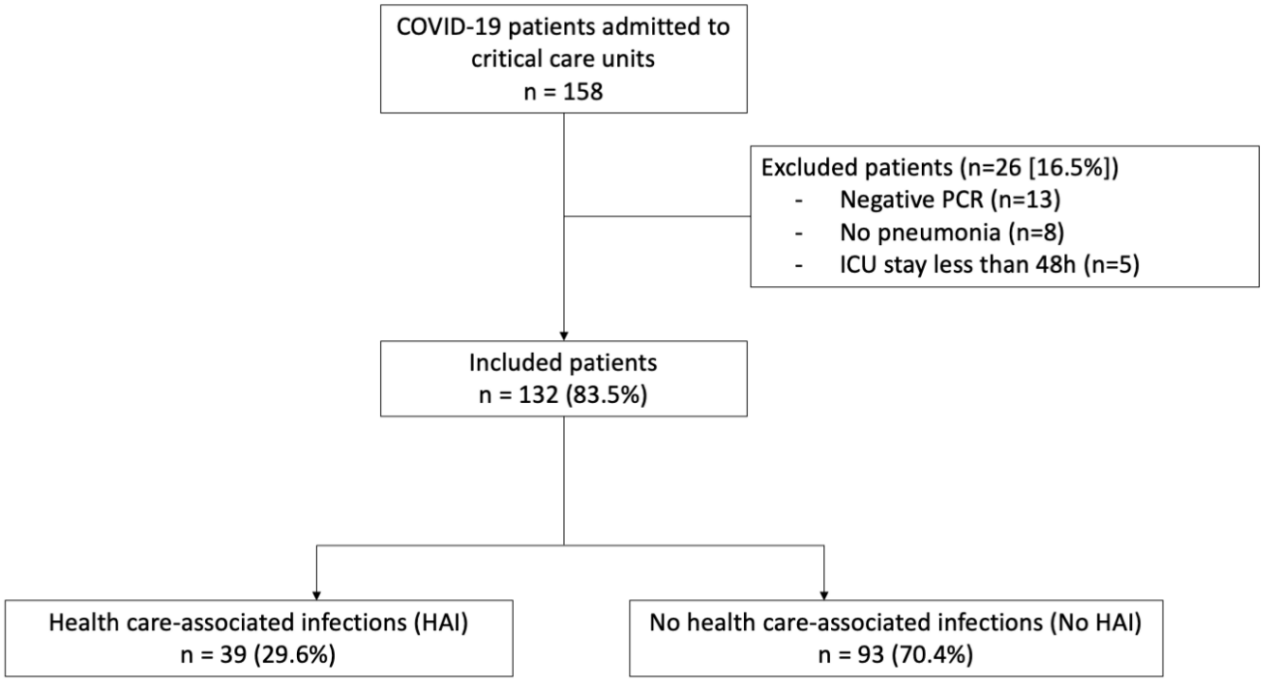

Figure 2e. Number of ICUs occupied per patient during admission to the COVID critical care area (**Figure 2eA**). Patient movement (static or no bed transfers, imported and exported patients) in each unit (**Figure 2eB**). Patient movement among units (**Figure 2eC**).

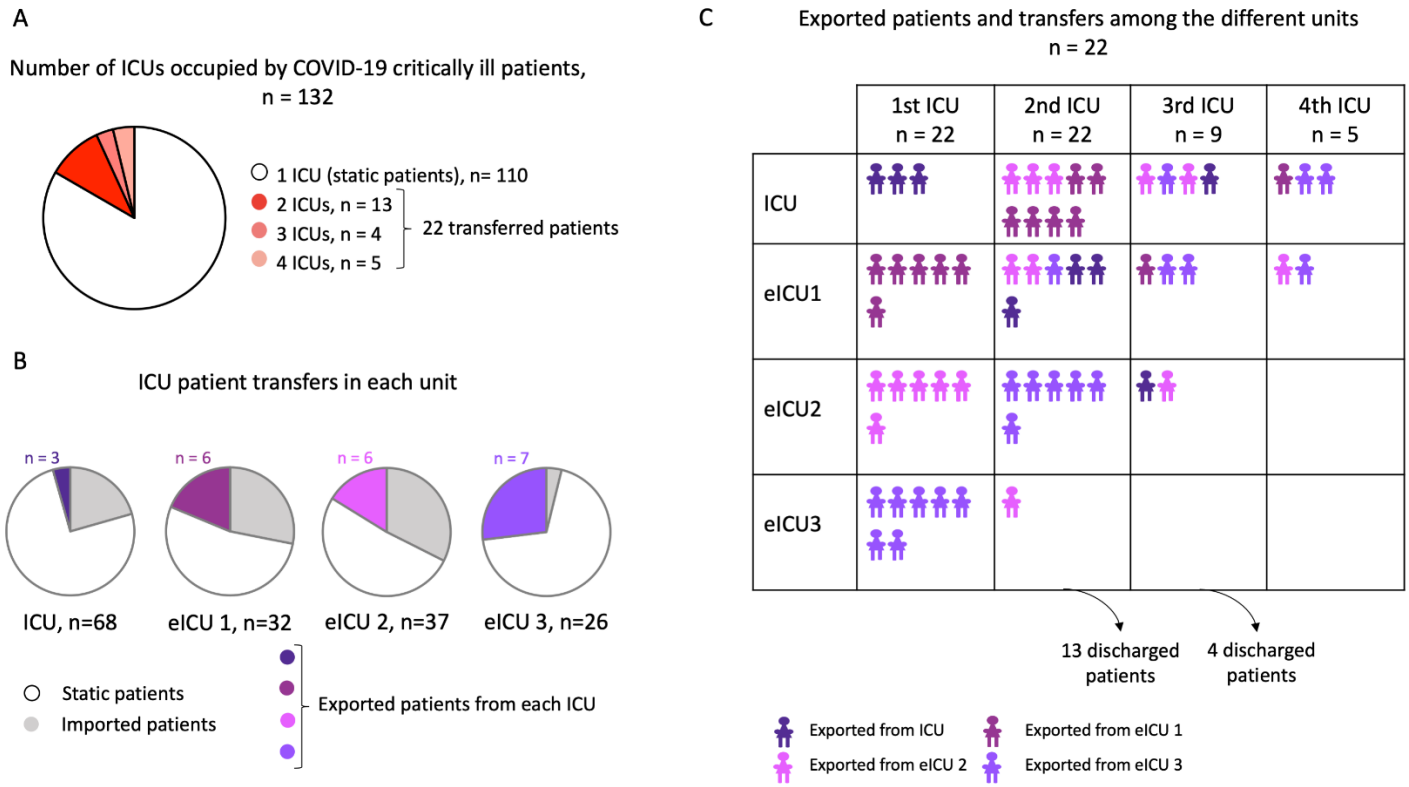

Figure 3e. Rates of HAIs in patients with COVID-19 pneumonia in the different COVID-19 intensive care units.

Footnote Figure 3e: HAI: health care-associated infection; BACT: bacteraemia; CRBSI: catheter-related bloodstream infection; BUO: bacteraemia of unknown origin; SB: secondary bacteraemia; UTI: urinary tract infection; VLRTI: ventilator-associated lower respiratory tract infection; VAT: ventilator-associated tracheobronchitis; VAP: ventilator-associated pneumonia; IPU: infected pressure ulcer; TSI: tracheostomy site infection.

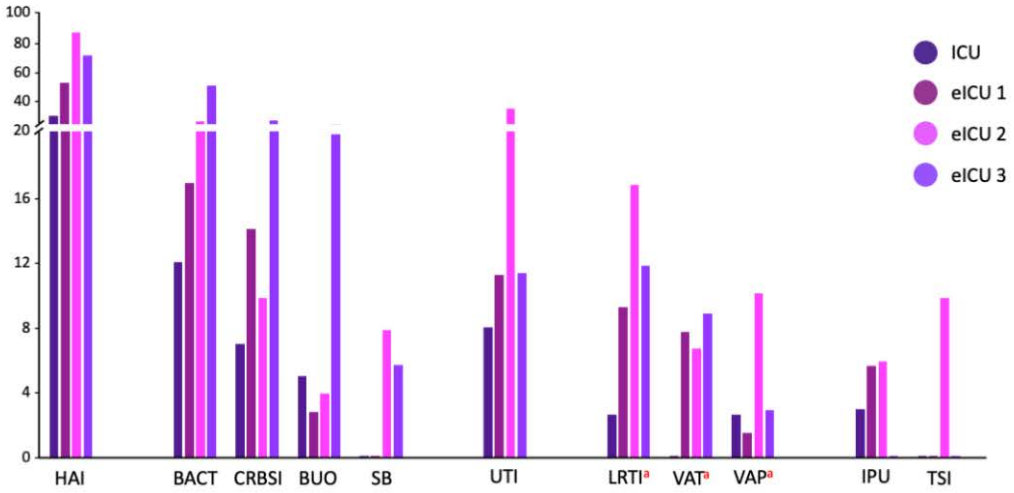

Figure 4e. Distribution over time of type of HAI in the COVID-19 intensive care units, overall (**Figure 4Ae**) and by each unit (**Figure 4Be**). Distribution over time of the microorganisms identified in each unit and type of HAI (**Figure 4Ce**).

Footnote Figure 4e: HAI: health-care associated infection; BACT: bacteraemia; UTI: urinary tract infection; VLRTI: ventilator-associated lower respiratory tract infection; IPU: infected pressure ulcer; TSI: tracheostomy site infection.

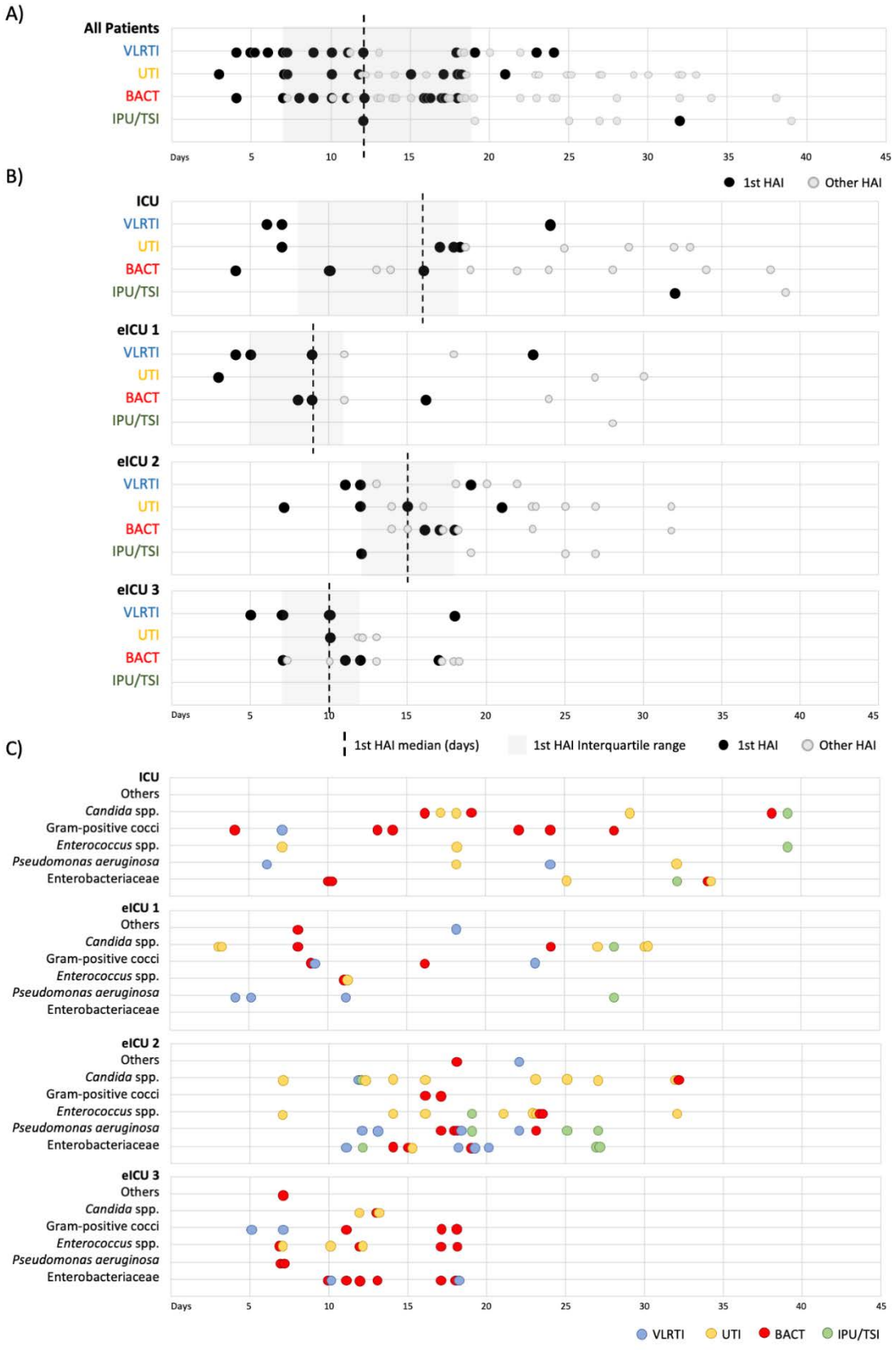

Figure 5e. Prevalence of microbiological results in the different HAIs.

Other included *Achromobacter xilosydians, Bacteroides fragilis, Bacteroides ovalus, Stenotrophomonas maltophilia, Aspergillus* spp, *Acinetobacter bereziniae; Candida* spp. included *Candida parapsilosis, Candida albicans, Candida tropicalis, Candida glabrata, Candida orthopsilosis.* Gram-positive cocci included *Staphylococcus aureus, Staphylococcus epidermidis, Streptococcus anginosus, Streptococcus agalactiae, Streptococcus pyogenes. Enterococcus* spp. included *Enterococcus faecalis* and *Enterococcus faecium. Enterobacteriaceae* included *Klebsiella pneumoniae, Enterobacter aerogenes, Escherichia coli, Serratia marcescens, Klebsiella oxytoca, Hafnia alvei, Citrobacter freundii, Citrobacter braakii, Enterobacte*r cloacae, *Citrobacter farmeri*.

Footnote Figure 5e: HAI: health care-associated infection; BACT: bacteraemia; CRBSI: catheter related bloodstream infection; BUO: bacteraemia of unknown origin; SB: secondary bacteraemia; UTI: urinary tract infection; VLRTI: ventilator-associated lower respiratory tract infection; VAT: ventilator-associated tracheobronchitis; VAP: ventilator-associated pneumonia; IPU: infected pressure ulcer; TSI: tracheostomy site infection.

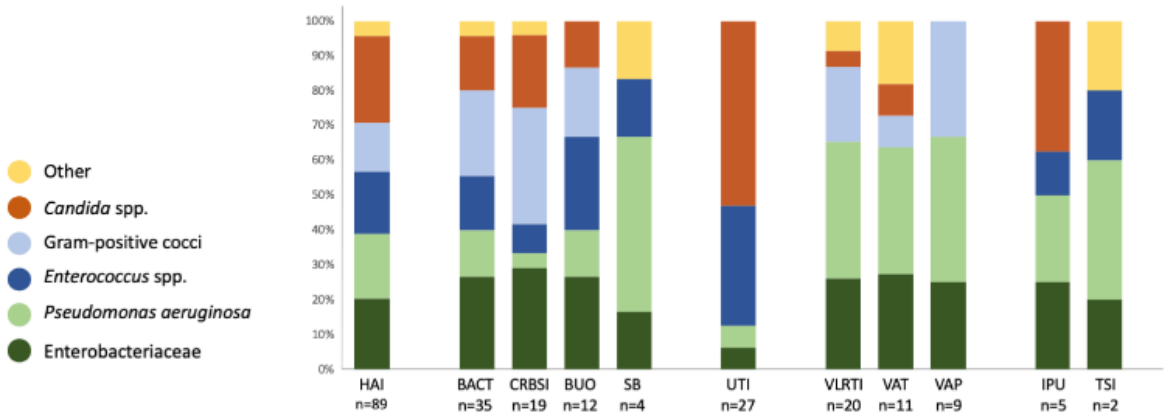

Figure 6e. Number and type of HAIs in survivors and non-survivors over time.

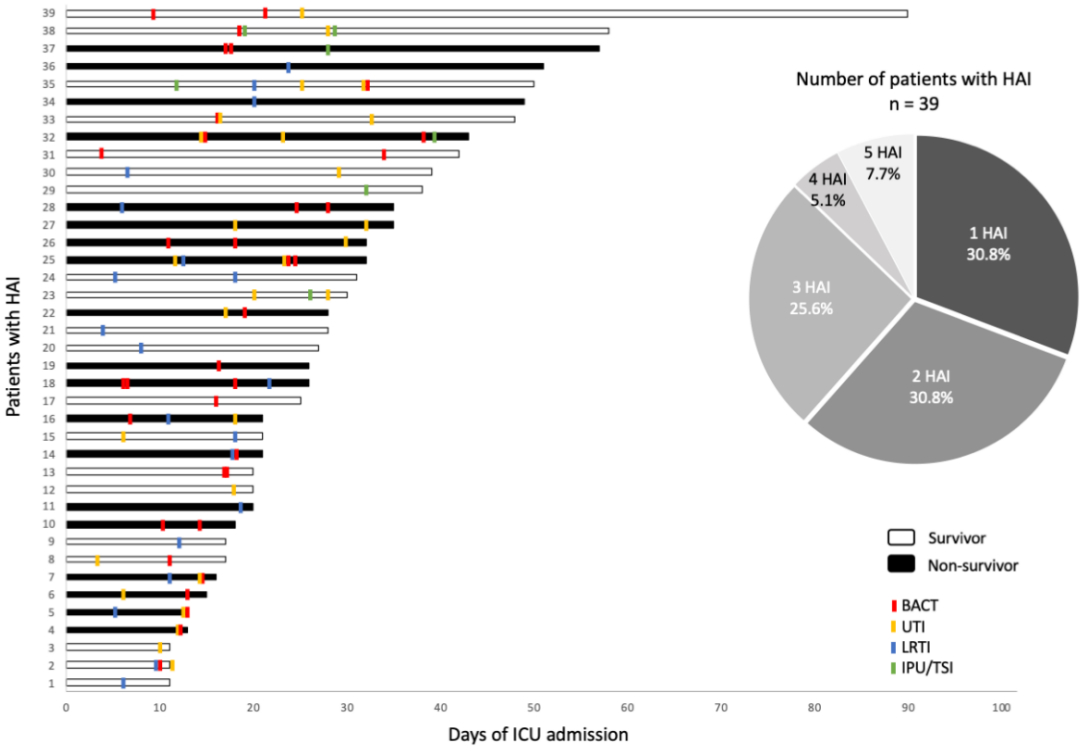

Table 1e. Characteristics of hospitalized COVID-19 patients by presence of health care-associated infection.

	No HAI n = 93	HAI n = 39	p-value
Demographics			
Age, years	60 (14)	65 (9)	0.012
Gender, male	60 (64.5)	24 (61.5)	0.746
Body mass index (Kg/m^2)	29 (5)	32 (6)	0.081
APACHE II score	14 (6)	17 (8)	0.018
Chronic comorbidities	67 (72.0)	32 (82.1)	0.226
Chronic lung disease			
Asthma	7 (7.5)	0 (0.0)	0.078
COPD	11 (11.8)	4 (10.3)	0.795
Cardiovascular disease			
Coronary artery disease	4 (4.3)	4 (10.3)	0.191
Hypertension	44 (47.3)	25 (64.1)	0.078
Atrial fibrillation	9 (9.7)	5 (12.8)	0.593
Neurological disease			
Stroke	0 (0.0)	3 (7.7)	0.007
Other medical conditions			
Immunosuppression	7 (7.5)	3 (7.7)	0.974
Alcoholism	3 (3.2)	4 (10.2)	0.029
Current or former smoker	26 (28.0)	12 (31)	0.648
Dislipidaemia	29 (31.2)	14 (35.9)	0.598
Diabetes mellitus	18 (19.4)	16 (41.0)	0.009
Liver disease	3 (3.2)	0 (0.0)	0.257
Chronic renal failure	6 (6.5)	3 (7.7)	0.796
Haematological malignancies	4 (4.3)	1 (2.6)	0.631
Solid tumour	12 (13.0)	7 (17.9)	0.466
Hypothyroidism	4 (4.3)	4 (10.3)	0.191
Chronic medications			
Inhaled corticosteroids	12 (12.9)	3 (7.7)	0.389
Chemotherapy	1 (1.1)	0 (0.0)	0.516
Biological drugs	2 (2.2)	0 (0.0)	0.363
ACE inhibitors	19 (20.4)	8 (20.5)	0.991
Angiotensin II blockers	10 (10.8)	4 (10.3)	0.933
Statins	19 (20.4)	13 (33.3)	0.115
Oral corticosteroids	5 (5.4)	2 (5.1)	0.954
Analytical variables			
C-reactive protein, mg/dL	14.6 (6.9-22.1)	16.8 (6.5-25.6)	0.465
Procalcitonin, ng/mL	0.24 (0.13-0.45)	0.21 (0.11-0.42)	0.800
Lymphocytes, x10^3/uL	0.81 (0.61-1.09)	0.82 (0.63-1.20)	0.950
IL-6, pg/mL	64.8 (34.8-120.7)	118.0 (73.3-139.2)	0.290
Ferritin, ng/mL	1099 (688-2429)	674.5 (446-2444)	0.244
Fibrinogen, mg/dL	500 (500-500)	500 (500-500)	0.995
D-dimer, ng/mL	855 (470-1770)	910 (522-1480)	0.986
Respiratory variables			
PaO_2 / FiO_2 ratio	119 (79-165)	92 (66-134)	0.396
Mechanical ventilation	53 (57.0)	39 (100)	<0.001
Days on mechanical ventilation	11 (6-19)	24 (14-33)	<0.001
Prone position	38 (40.9)	29 (74.4)	<0.001
Tracheostomy	16 (17.2)	27 (69.2)	<0.001

Other interventions			
Renal replacement therapy	4 (4.3)	12 (30.8)	<0.001
Admission to more than 1 ICU	6 (6.5)	16 (41.0)	<0.001
Pharmacological treatment			
Neuromuscular blockers	19 (20.4)	24 (61.5)	<0.001
Hydroxychloroquine	92 (98.9)	37 (94.9)	0.154
Antivirals	28 (30.1)	11 (28.2)	0.827
Immunomodulators	19 (20.4)	2 (5.1)	0.028
Antimicrobials	83 (89.2)	39 (100)	0.033
DOT of antimicrobials	113 (60-163)	180 (115-218)	<0.001
Vasopressors	47 (50.5)	39 (100)	<0.001
Systemic corticosteroids	86 (92.5)	36 (92.3)	0.974
Use of SDD	34 (36.6)	20 (51.3)	0.116
Complications			
ARDS	81 (88.0)	37 (94.9)	0.232
Pulmonary embolism	4 (18.2)	1 (9.1)	0.492
Cardiac arrest	3 (3.2)	2 (5.1)	0.601
Stroke	1 (2.7)	1 (5.3)	0.625
Arrhythmia	7 (7.5)	5 (12.8)	0.334

Data expressed as frequencies and percentages [n (%)] or mean (SD) or medians and interquartile ranges (IQR or 25th-75th percentile).
APACHE II: acute physiology and chronic health evaluation II; COPD: chronic obstructive pulmonary disease; SDD: selective decontamination of the digestive tract; DOT: days of treatment; IL-6: interleukin 6; ARDS: acute respiratory distress syndrome, ICU: intensive care unit.

Table2e. Etiology of the different types of health care-associated infections (HAI) diagnosed in COVID-19 patients.

	CRBSI	BUO	SB	UTI	VLRTI	IPU	TSI	Total
GPC	**11 (44)**	**7 (47)**	**1 (17)**	**11 (34)**	**5 (22)**	**1 (13)**	**1 (20)**	**37 (32)**
S aureus								
Susceptible	1 (4)	2 (13)	-	-	3 (13)	-	-	6 (5)
Resistente a meticilina	-	-	-	-	1 (4)	-	-	1 (1)
S epidermidis	7 (28)	1 (7)	-	-	-	-	-	8 (7)
E faecalis	1 (4)	3 (20)	-	7 (22)	-	-	-	11 (10)
E faecium	1 (4)	1 (7)	1 (17)	4 (13)	-	1 (13)	1 (20)	9 (8)
S agalactiae	-	-	-	-	1 (4)	-	-	1 (1)
S anginosus	1 (4)	-	-	-	-	-	-	1 (1)
GNB	**9 (36)**	**6 (40)**	**5 (83)**	**4 (13)**	**17**	**4 (50)**	**4 (80)**	**49 (43)**
E coli								
Susceptible	-	-	-	-	-	1 (13)	-	1 (1)
ESBL	-	1 (7)	1 (17)	2 (6)	2 (9)	-	-	6 (5)
K pneumoniae								
Susceptible	-	1 (7)	-	-	1 (4)	-	1 (20)	3 (3)
ESBL	5 (20)	1 (7)	-	-	2 (9)	-	-	8 (7)
E aerogenes	1 (4)	-	-	-	-	-	-	1 (1)
S marcescens	-	1 (7)	-	-	1 (4)	1 (13)	-	3 (3)
Hafnia alvei	1 (4)	-	-	-	-	-	-	1 (1)
P aeruginosas								
Susceptible	1 (4)	2 (13)	3 (50)	-	8 (35)	2 (25)	2 (40)	18 (16)
MDR	-	-	-	2 (6)	1 (4)	-	-	3 (3)
Acinetobacter bereziniae	1 (4)	-	-	-	-	-	-	1 (1)
A xilosydans	-	-	1 (17)	-	2 (9)	-	-	3 (3)
B fragilis	-	-	-	-	-	-	1 (20)	1 (1)
Fungus	**5 (20)**	**2 (13)**	**0 (0)**	**17 (53)**	**1 (4)**	**3 (38)**	**0 (0)**	**28 (25)**
C albicans	3 (12)	-	-	10 (31)	-	1 (13)	-	14 (12)
C parapsilopsis								
Susceptible	-	1 (7)	-	-	1 (4)	-	-	2 (2)
Fluconazole-resistant	1 (4)	1 (7)	-	-	-	1 (13)	-	3 (3)
C tropicalis	1 (4)	-	-	2 (6)	-	1 (13)	-	4 (4)
C glabrata	-	-	-	3 (9)	-	-	-	3 (3)
Candida spp	-	-	-	1 (3)	-	-	-	1 (1)
C orthopsilosis	-	-	-	1 (3)	-	-	-	1 (1)
TOTAL	**25 (100)**	**15 (100)**	**6 (100)**	**32 (100)**	**23 (100)**	**8 (100)**	**5 (100)**	**114 (100)**

Data expressed as frequencies and percentages [n (%)]. MDR: multidrug resistant; ESBL: extended-spectrum beta lactamases; CRBSI: catheter-related bloodstream infections; BUO: bacteremia of unknown origin; SB: secondary bacteremia; UTI: urinary tract infection; VLRTI: ventilator-associated lower respiratory tract infections; VAT: ventilator-associated tracheobronchitis; VAP: ventilator-associated pneumonia; IPU: infected pressure ulcers; TSI: tracheostomy site infection.

Table 3e. Patients that died in the first 72 hours after the diagnosis of HAI.
BC: blood cultures; PTC: plugged telescopic catheter; U: urine; TE: tracheal exudate

Patient	**1**	**2**	**3**	**4**	**5**	**6**
Date of eICU admission	29 Mar 2020	29 Mar 2020	28 Mar 2020	24 Mar 2020	20 Mar 2020	22 Mar 2020
Type of eICU	eICU 2	eICU 3	eICU 3	eICU 3	eICU 2	eICU 2
Date of HAI	15 Apr 2020	10 Apr 2020	10 Apr 2020	12 Apr 2020	08 Apr 2020	29 Apr 2020
Date of death	17 Apr 2020	11 Apr 2020	10 Apr 2020	14 Apr 2020	09 Apr 2020	04 May 2020
Type of HAI	VLRTI and UTI	BUO and UTI	CRBSI and UTI	CRBSI and VLRTI	VLRTI	TSI
Microbiologic aetiology of HAI and obtained sample	*E coli* ESBL (BC and PTC) *E faecium* + *C albicans* (U)	*K pneumoniae* ESBL + *E faecalis* (BC)	*K pneumoniae* ESBL + *C albicans* (BC) *C albicans* (U)	*K pneumoniae* ESBL + *E faecalis* (BC) *K pneumoniae* ESBL (PTC)	*E coli* ESBL (PTC)	*C parapsilosis* (BC) *C parapsilosis* + *E faecium* (TE)
Increased inflammatory pattern	Yes	No	Yes	Yes	No	Yes
Renal replacement therapy	No	No	No	Yes	Yes	Yes
Vasopressors	Yes	Yes	Yes	Yes	Yes	Yes
P_aO_2/F_iO_2 ratio (day of HAI)	100	52	138	177	175	177
Empirical treatment and start date	Meropenem + linezolid 14 Apr 2020	Meropenem + linezolid + anidulafungin 10 Apr 2020	Meropenem 10 Apr 2020	Meropenem + linezolid + anidulafungin 11 Apr 2020	No	Liposomal amphotericin B 26 Apr 2020
Previous antibiotic treatment (previous 72 hours)	No	No	Yes (linezolid)	No	No	Yes (daptomycin, meropenem and linezolid)
Appropriate previous treatment	-	-	No	-	-	No