Journal of
Anesthesia & Intensive Care Medicine
ISSN: 2474-7653

Juniper PUBLISHERS key to the Researchers

Short Communication
Volume 11 Issue 2 - March 2021
DOI: 10.19080/JAICM.2021.11.555809

J Anest & Inten Care Med

Bacterial and Fungal Infections in Critically Ill Patients with Covid-19 Pneumonia: An Observational Study

Rafael León[1] MD, Jorge Rodríguez-Gómez[1]* PhD, Sheila Cárcel[1] MS, Cristina López[1] MD, María Rojas-Amezcua[2] MD, Francisco Rivera-Espinar[3] MD, Ana Mula[1] MD, Marina Rodríguez[1] MD, Cristina Riazzo[4] MS, Juan Jesús Pineda-Capitán[1] MS Juan Carlos Pozo-Laderas[1] PhD, Julián De la Torre-Cisneros[5†] PhD, Carmen De la Fuente-Martos[1] MD

[1]*Intensive Care Unit. Reina Sofia Universitiy Hospital-Maimonides Biomedical Research Institute of Córdoba (IMIBIC)-University of Cordoba, Cordoba, Spain*

[2]*Intensive Care Unit. Hospital Infanta Margarita. Cabra, Spain*

[3]*Intensive Care Unit. Hospital de Montilla. Montilla, Spain*

[4]*Microbiology Unit, Reina Sofia University Hospital, Spain*

[5]*Infectious Diseases Unit. Reina Sofia University Hospital- Maimonides Biomedical Research Institute of Córdoba (IMIBIC)-University of Cordoba, Cordoba, Spain*

Submission: March 17, 2021; **Published:** March 30, 2021

***Corresponding author:** Jorge Rodríguez Gómez, PhD, Intensive Care Unit. Hospital Universitario Reina Sofia- Av Menendez Pidal s/n, 14004. Córdoba (Córdoba). España, Spain

[†]*Authors who are members of the Spanish Network for Research in Infectious Diseases (REIPI RD16/0016/0008/ REIPI RD16/0016/0001).*

Abstract

Objectives: The aim of this study is describe and analyse bacterial and/or fungal infections in critically ill patients with coronavirus diseases 2019 (COVID-19) pneumonia during the first pandemic period and compared them with historic cohort of influenza pneumonia patients.

Design: Multicentre, observational, and retrospective.

Setting: Three ICUs of Spain

Patients: Critically ill patients with COVID-19 and influenza pneumonia which required ICU admission were recruited from October 2018 to April 30th, 2020. We described population, distribution of infections/microorganisms and risk factors related.

Main variables of interest: Exposure: COVID-19 infection. Outcome: Superinfection, Overall mortality 90 days

Measurements and Main Results: 58 critical care patients with COVID-19 and a further 48 patients with influenza pneumonia were included. COVID-19 patients had significantly less respiratory co-infection (3 vs 29; $p<0.001$). Superinfections were not significantly higher in patients with COVID-19 (46 vs 29%; $p=0.06$). Gram negative bacteria were significantly higher than in influenza pneumonia patients (48 vs 27; $p=0.02$) and *Pseudomonas aeruginosa* (27%), and lower respiratory infections (42 vs 25; $p=0.07$) were the most frequent aetiology and source of superinfection in COVID-19 patients. In adjusted analysis, days of invasive mechanical ventilation (OR 1.08; CI 1.01-1.16; $p=0.04$) and SOFA score at admission (OR 1.48; CI 1.13-1.95; $p=0.04$) were statistically associated with superinfection. Superinfection (HR 4.76; CI 1.63-13.91; $p=0.004$) was related in crude analysis with overall mortality at 90 days.

Conclusion: The incidence of coinfections is low in critically ill patients with COVID-19 pneumonia and significantly less frequent than in patients with influenza pneumonia. Empirical treatment at admission should be closely revaluated. Incidence of superinfection is high, if is suspected, empirical antipseudomonal treatment should be considered specially in lower respiratory infections, considering local flora. Severity of illness and supportive treatments seems to be the *clearer* factors related to superinfections.

Keywords: Coronavirus infection, Pneumonia, Infection, Coinfection, Superinfection, Critical care

Introduction

Information about bacterial and fungal infections in critical patients with Coronavirus disease 2019 (COVID-19) is increasing after one year of the pandemic outbreak. Some explanations of this fact are the most immediate priorities for hospitals providing acute medical care (intensivist have habilitated up to 300% more critical patients' beds in hospitals), keeping critically ill patients alive, protecting staff, and optimizing care of critically ill non-COVID patients in this context [1-3].

Coinfection in patients with COVID-19 seems to be low according to published data, but complications with superinfections in these critically ill patients shows important variability in different studies [4]. While some reports show low incidence of hospital acquired pneumonia of 11%, others show an increase of incidence of more than 50% of lower respiratory infections or high incidence of blood stream infections in these patients [5,6].

The aim of this study is to describe bacterial and/or fungal coinfections and superinfections in critically ill patients with COVID-19 pneumonia, compare them with critical care patients with influenza pneumonia and evaluate the risk factors associated. This knowledge may be useful to improve management of these patients, optimize antibiotics policy and to adapt initial protocols.

Patients and Methods

This is an observational, retrospective, cohort study carried out at 3 hospitals in the south of Spain. We evaluated all adult patients with viral pneumonia due to SARS-VoV-2 and influenza who required ICU admission from October 2018 to April 30th, 2020. Eligibility criteria included were age ≥18 years old, confirmed laboratory test of SARS-COV-2 or influenza (A or B) infection, and compatible chest X ray or computed tomography infiltrates. Patients with limited therapeutic effort at admission and ICU hospitalization duration lasting <48 hours were excluded.

Supplementary Table 1: Checklist of items according to STROBE document.

Title and abstract	Indicate the study design with a commonly used term in the title or abstract.	Study design indicated in title or abstract with a commonly used term
	Provide an informative and balanced summary in the abstract of what was done and what was found	Balanced and informative summary included in abstract
Background/ rationale	Explain the scientific background and rationale for the investigation being reported	The scientific background and rationale are explained in the Introduction
Objectives	State specific objectives, including any pre-specified hypotheses	Hypothesis and objectives included in the Introduction
Study design	Present key elements of study design early in the paper	Study design included in Methods
Setting	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	Included in Methods
Participants	Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up	Included in Methods
	For matched studies, give matching criteria and number of exposed and unexposed	Not a matched study
Variables	Clearly define all outcomes, exposures, predictors,	All included in Methods
	potential confounders, and effect modifiers. Give	
	diagnostic criteria, if applicable	
Data sources/ measurement	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	Specified in Methods. The same data collection method was used in the groups.
Bias	Describe any efforts to address potential sources of bias	Selection bias: inclusion of consecutive cases. Information bias: use of standard and well-defined variables. For risk factors associated we applied logistic regression
Study size	Explain how the study size was arrived at	Consecutive cases from October 2018 to April 2020 of COVID-19 and Influenza patients

How to cite this article: Rafael L M, Jorge R-G P, Sheila C M, Cristina L M, María Rojas-A. Bacterial and Fungal Infections in Critically Ill Patients with Covid-19 Pneumonia: An Observational Study. J Anest & Inten care med. 2021; 11(2): 555809. DOI:10.19080/JAICM.2021.11.555809.

Quantitative variables	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why	Quantitative variables were handled as quantitative described in median and interquartile range to avoid extreme results
Statistical methods	Describe all statistical methods, including those used to control for confounding	Included in Methods. Logistic regression
	Describe any methods used to examine subgroups and interactions	In multivariable analysis we avoid to applied similar variables. Variance inflaction factor was used to check multicollinearity
	Explain how missing data were addressed	Missing data were not included
	If applicable, explain how loss to follow-up was addressed	No patient was lost
	Describe any sensitivity analyses	Included in Methods
Participants	Give characteristics of study participants (e.g., demographic, clinical, social) and information on exposures and potential confounders	Table 1
	Indicate number of participants with missing data or each variable of interest	Patient included and excluded described in results
	Summarise follow-up time (e.g., average, and total amount)	In variable length of stay in ICU
Descriptive data	Report numbers of outcome events or summary measures over time	Table 1
Main results	Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	Specified in Results and table 2
	Report category boundaries when continuous variables were categorized	Specified in table 1.
	If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period.	Not applicable
Outcome data	Report numbers of outcome events or summary measures over time	Table 1 and 2
Other analyses	Report other analyses done—e.g., analyses of subgroups and interactions, and sensitivity analyses	Specified in table 1, 2, 3 and results
Key results	Summarise key results with reference to study objectives	Specified in Research into context, Abstract and Discussion
Limitations	Discuss limitations of the study, considering sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias	Included in Discussion
Interpretation	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	Included in Discussion
Generalisability	Discuss the generalisability (external validity) of the study results	Included in Discussion taking into in account low sample
Funding	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	No funding. Collaboration of authors of REIPI network

How to cite this article: Rafael L M, Jorge R-G P, Sheila C M, Cristina L M, María Rojas-A. Bacterial and Fungal Infections in Critically Ill Patients with Covid-19 Pneumonia: An Observational Study. J Anest & Inten care med. 2021; 11(2): 555809. DOI:10.19080/JAICM.2021.11.555809.

The study was approved by the local Ethics Committee of the Reina Sofía University Hospital of Córdoba (Code 4706), which were exempted from the need to seek written informed consent due to the observational and retrospective nature of the study. STROBE recommendations for observational studies were followed and checked (Supplementary Table 1) [7]

Clinical data was recorded in a standardized protocol and anonymized in a database. Variables evaluated were: (i) demographics, (ii) previous conditions, (iii) clinical characteristics, (iv) severity of illness (APACHE II and SOFA score) (v) analytic and microbiological studies, (vii) pharmacologic and support therapy, (viii) and outcome variables (microbiologically confirmed coinfection and superinfection during ICU stay, type of infection, microbiology characteristics, overall mortality at 30 and 90 days and died with active infection). Corticosteroids when administered were given at maximum doses of 1mg/kg/day of methylprednisolone. All patients admitted during this period were followed until ICU discharge and overall mortality 90 day was assessed by phone call in discharge patients.

To standardize the classification of an infection as present on admission (co-infection) or a healthcare-associated infection (superinfection), we used the CDC/NHSN criteria. Superinfection is defined as localized or systemic condition resulting from an adverse reaction to the presence of an infectious agent(s) or its toxin(s) that was not present on or after the 3rd hospital day (day of hospital admission is day 1) [8]. Diagnosis of lower respiratory infection was based on two of the next criteria: temperature up to 38.5 °C or leucocyte account (≥12.000 or <4.000 cells per µL), and purulent secretions. Episodes need microbiological confirmation with isolation in the endotracheal aspirate. Ventilator associated pneumonia was defined with the above criteria and the presence of new or progressive infiltrates on radiological explorations after 48 hours of mechanical ventilation [6]. ICU bloodstream infection for typical skin contaminants (e.g. coagulase-negative Staphylococcus) were included only if ≥2 blood cultures showed the same phenotype on separate occasions, or 1≥ blood cultures for clinical sepsis and no other infectious process; catheter related bloodstream infections were documented by quantitative tip culture [9]. All episodes and criteria were critically reviewed by two investigators (A.M and J.R.G).

SARS-COV-2 testing was performed at Microbiology Reference Laboratory (Reina Sofía University Hospital/IMIBIC). VIASURE SARS-CoV-2 Real Time PCR Detection Kit (CerTest Biotec), a multiplex real-time PCR detecting ORF1ab and N genes, was used for diagnosis. Influenza A/B virus were also investigated using GeneXpert® (Cepheid) or the BioFire® FilmArray® Respiratory 2 plus Panel (Biomerieux).

Continuous variables were compared using the Mann-Whitney U or T Test of Student Test and categorical variables were compared using the Chi Square Test or Fisher's Exact Test as appropriate. Ventilator associated pneumonia and blood stream infections incidence rate were calculated as the number of events per 1000 patient-days at risk; the 95% confidence interval (CI) for the incidence rate estimate was obtained using mid-P exact test. In patients with COVID-19, multivariate analyses were performed using logistic regression for study variables related with superinfection and Cox regression for overall mortality 90 days. In logistic regression for variables related to superinfection, each multivariate analysis included one variable for every ten events; they were selected those with p<0.1 (model 1) or those with clinical interest (model 2 and 3). Due to prolonged length of ICU stays of COVID-19 patients we decide overall 90 days mortality over overall 30 mortality. In Cox regression analysis for overall mortality 90 days, superinfection variable was included as time-dependent; patients have an additional transient state before superinfection event, not taking timing, this time, into account would cause time-depending bias [10].

Results

Description of patients with COVID19 and Influenza pneumonia

During the study period 116 patients were evaluated. 62 patients with highly suspected COVID-19 pneumonia were identified; 58 met inclusion criteria, 3 patients with COVID-19 disease like symptoms were not confirmed in laboratory RT-PCR of SARS-COV-2 and 1 patient with limited therapeutic effort at admission was excluded. There were 48 influenza pneumonia patients evaluated and all of them were legible without exclusion criteria.

Clinical characteristics of COVID-19 and influenza pneumonia cohorts are described in Table 1. Critically ill patients with COVID-19 pneumonia were median age 64 years old. Only 10% (6) of patients did not have comorbidities while the most frequent were hypertension and obesity with 56% (33) and 37% (22) respectively. At admission, laboratory results showed the median of D Dimer of 1096 ng/ml, median of 347 cell/ **µ**L of CD4 and 47 pg/mL of IL6.

Patients with COVID-19 in comparison with those with influenza pneumonia were significantly older (64 vs 55; p=0.004), with higher incidence of hypertension (56 vs 37; p=0.04), more days from onset of symptoms to ICU admission (9 vs 4; p<0.001), treated with corticosteroids (43 vs 16%; p=0.002), but have less APACHE II score (12 vs 19; p<0.001).

Co-infection and superinfections

Patients with COVID-19 pneumonia, in comparison, had significantly less frequent respiratory co-infection (3 vs 29; p<0.001) (Table 2). 96% of patients with COVID-19 were treated with antibiotics at admission. There were 3 cases of Coagulase Negative Staphylococcus in COVID-19 patients at ICU admission.

How to cite this article: Rafael L M, Jorge R-G P, Sheila C M, Cristina L M, María Rojas-A. Bacterial and Fungal Infections in Critically Ill Patients with Covid-19 Pneumonia: An Observational Study. J Anest & Inten care med. 2021; 11(2): 555809. DOI:10.19080/JAICM.2021.11.555809.

Table 1: Clinical characteristics of patients with viral pneumonia, COVID-19 or influenza, admitted in ICU. All data are expressed as n (%) unless otherwise indicated; COVID-19: Coronavirus disease 2019; ICU: Intensive care unit; IQR: interquartile range; BMI: Body mass index; APACHE II score: Acute physiology and chronic health evaluation II; SOFA score: Sequential Organ Failure Assessment; CRP: C-Reactive protein; PCT: Procalcitonin; IL6: Interleukin 6; ECMO: Extracorporeal membrane oxygenation; NA: Not available.

Variables	Influenza	COVID-19	*p*
	(n=48)	(n=58)	
Demographics			
Age, median (IQR)	53 (44-65)	64 (53-73)	**0.003**
Male gender	31 (64.6)	36 (62.1)	0.78
Previous conditions			
Underlying conditions			
Without comorbidities	6 (12.5)	6 (10.3)	0.72
Obesity (BMI > 30)	14 (29.2)	22 (37.9)	0.34
Hypertension	18 (37.5)	33 (56.9)	**0.04**
Diabetes mellitus	11 (22.9)	16 (27.6)	0.58
Chronic heart disease	10 (20.8)	5 (8.6)	0.07
Chronic respiratory disease	11 (22.9)	10 (17.2)	0.46
Chronic renal failure	2 (4.2)	4 (6.9)	0.54
Oncologic conditions	7 (14.6)	6 (10.3)	0.5
Autoimmune disorders	2 (4.2)	3 (5.2)	1
Solid organ transplantation	0 (0)	2 (3.4)	0.5
Charlson comorbidity index, median (IQR)	0 (1-3)	3 (1-4)	0.07
Surgery previous three months	0 (0)	4 (6.9)	0.12
Clinical characteristics and severity			
Days from onset symptoms to ICU admission (IQR)	4 (3-7)	9.5 (7-13)	**<0.001**
ICU length of stay, median (IQR)	11 (5-18)	8.5 (5-34)	0.97
Severity			
APACHE II score, median (IQR)	19 (14-23)	12 (8.5-16.5)	**<0.001**
SOFA score at admission, median (IQR)	5 (3-8)	3 (3-7)	0.24
SOFA score at 72 hours, median (IQR)	4 (2-8)	5 (3-7.5)	0.85
Laboratory findings at ICU admission			
Total leucocytes (10^9/L), median (IQR)	8.75(4.24-13.92)	8.20(5.58-10.58)	0.87
Neutrophils (%), median (IQR)	85 (76.5-87-5)	86 (81-90)	0.15
Lymphocytes (10^9/L), median (IQR)	0.5 (0.37-0.84)	0.65 (0.36-1.01)	0.61
CD4 Lymphocyte (cell/µL, n=31), median (IQR)	NA	347 (248-476)	NA
D Dimer (ng/mL), median (IQR)	866 (545-2537)	1096 (646-3991)	0.61
CRP (mg/L), median (IQR)	291 (163-292)	280 (76-295)	0.92
PCT (ng/mL), median (IQR)	1.12 (0.6-5.5)	0.18 (0.08-0-52)	0.24
IL 6 (pg/mL, n=35), median (IQR)	NA	47 (14-129)	NA
Treatment			

How to cite this article: Rafael L M, Jorge R-G P, Sheila C M, Cristina L M, María Rojas-A. Bacterial and Fungal Infections in Critically Ill Patients with Covid-19 Pneumonia: An Observational Study. J Anest & Inten care med. 2021; 11(2): 555809. DOI:10.19080/JAICM.2021.11.555809.

Invasive mechanical ventilation	31 (64.6)	39 (67.2)	0.77
Length of invasive mechanical ventilation, median (IQR)	7 (0-16)	8 (0-28)	0.28
Tracheostomy	6 (12.5)	15 (25.9)	0.08
Vasoconstrictor agents	31 (64.6)	38 (65.5)	1
Renal replacement therapy	9 (18.8)	9 (15.5)	0.65
ECMO	2 (4.2)	1 (1.7)	0.58
Antibiotic treatment at ICU admission	46 (95.8)	56 (96.6)	1
Antiviral agents	48 (100)	51 (89.5)	**0.03**
Immunomodulatory therapy	3 (6.3)	36 (62.1)	
Tociluzumab	0 (0)	17 (29.3)	NA
Corticosteroids	7 (14.6)	25 (43.1)	**0.001**
Non-specific immunoglobulin	0 (0)	11 (19)	NA
Outcome			
Microbiologically confirmed respiratory coinfection	14 (29.2)	2 (3.4)	**<0.001**
Microbiologically confirmed superinfection	14 (29.2)	27 (46.6)	0.06
Overall mortality 30 day	7 (14.6)	11 (19)	0.55
Overall mortality 90 day	9 (18.8)	17 (29.3)	0.2
Died with active infection	7 (14.6)	11 (19)	0.55

Table 2: Characteristics of microbiologically confirmed bacterial and/or fungal infections in patients with viral pneumonia, COVID-19 or influenza, admitted in ICU. All data are expressed as n (%) unless otherwise indicated; COVID-19: Coronavirus disease 2019; ICU: Intensive care unit; NA: Not available; CRP: C-Reactive protein; PCT: Procalcitonin.

MICROBIOLOGICALLY CONFIRMED COINFECTION	Influenza	COVID-19	*p*
	n=48	n=58	
Patients with microbiologically confirmed coinfection	14 (29.2)	5 (8.6)	**0.006**
Patients with microbiologically confirmed respiratory coinfection	14 (29.2)	2 (3.4)	**<0.001**
Positive urinary antigen test			
Positive *Legionella pneumoniae* urinary antigen test	0 (0)	0 (0)	**NA**
Positive *Streptococcus pneumoniae* urinary antigen test	5 (10.4)	1 (1.7)	**0.04**
Positive respiratory cultures	12 (25)	1 (1.7)	**<0.001**
Streptococcus pneumoniae	6 (12.5)	0 (0)	
Staphylococcus aureus	4 (8.3)	1 (1.7)	
Aspergillus spp	1 (2)	1 (1.7)	
Gram-negative bacteria	5 (10.4)	0	
Positive viral tests			NA
SARS CoV2	0 (0)	58 (100)	
Influenza A	44 (91.7)	0 (0)	
Influenza B	4 (8.3)	0 (0)	
Others[a]	2 (4.1)	0 (0)	
Positive blood cultures	2 (4.1)	3 (5.1)	0.39
Coagulase-negative Staphylococcus	0 (0)	3 (5.1)	

How to cite this article: Rafael L M, Jorge R-G P, Sheila C M, Cristina L M, María Rojas-A. Bacterial and Fungal Infections in Critically Ill Patients with Covid-19 Pneumonia: An Observational Study. J Anest & Inten care med. 2021; 11(2): 555809. DOI:10.19080/JAICM.2021.11.555809.

Others	2 (4.1)	2 (3.4)	
MICROBIOLOGICALLY CONFIRMED SUPERINFECTION	**Influenza**	**COVID-19**	***p***
Confirmed episodes			
Patients with microbiologically confirmed superinfection	14 (29.2)	27 (46.6)	0.06
Number of episodes of bacterial and/or fungal superinfection/patient	0 (0-1)	0 (0-1)	0.15
Type of infection	36	70	
Lower respiratory infections	9 (25)	30 (42.9)	0.07
Ventilator associated pneumonia	4 (11.1)	15 (21.4)	0.19
Other lower respiratory infections	5 (13.9)	15 (21.4)	0.34
ICU Blood stream superinfections	17 (47.2)	28 (40)	0.47
Unknow source blood stream infection	12 (33.3)	12 (17.1)	0.05
Intra-vascular catheter	5 (13.5)	16 (22.9)	0.27
Urinary tract infection	9 (25)	6 (8.6)	**0.02**
Others	1 (2.8)	6 (8.6)	0.41
Microorganisms isolated	44	98	
Gram-positive bacteria	24 (54.5)	42 (42.9)	0.19
Coagulase-negative Staphylococcus	12 (23.7)	17 (17.3)	
Enterococcus spp	12 (25)	22 (22.4)	
Staphylococcus aureus	0 (0)	2 (2)	
Streptococcus anginosus	1 (2.3)	1 (1)	
Gram-negative bacteria	12 (27.3)	47 (48)	**0.02**
Escherichia coli	0 (0)	7 (7.1)	
Pseudomonas aeruginosa	9 (20.7)	27 (27.6)	
Klebsiella oxytoca	0 (0)	2 (2)	
Klebsiella pneumoniae	2 (4)	3 (3)	
Others	1 (2.3)	8 (8.1)	
Fungi	8 (18.2)	9 (9.2)	0.12
Candida spp	8 (18.2)	8 (8.2)	
Aspergillus spp	0 (0)	1 (1)	
Resistance	n=44	n=98	
Non-multidrug-resistant	29 (62.9)	73 (74.5)	0.29
Multidrug-resistant	6 (13.6)	11 (11.2)	0.68
Extensively drug-resistant	3 (6.8)	8 (8.2)	0.78

[a]Others: respiratory syncytial virus (1), cythomegalovirus (1)

[b]Tracheobronchitis and 2 hospital acquired pneumonia in COVID-19 cohort.

Superinfections were not significantly higher in patients with COVID-19 (46 vs 29%; p=0.06). The most frequent microorganisms isolated in this cohort was *Pseudomonas aeruginosa* (27%), and Gram-negative bacteria were significantly higher than in influenza pneumonia patients (48 vs 27; p=0.02) (Table 2). Lower respiratory infections were the most frequent superinfections in COVID-19 patients, and they were not significantly higher than influenza pneumonia patients (42 vs 25; p=0.07); gram negative

How to cite this article: Rafael L M, Jorge R-G P, Sheila C M, Cristina L M, María Rojas-A. Bacterial and Fungal Infections in Critically Ill Patients with Covid-19 Pneumonia: An Observational Study. J Anest & Inten care med. 2021; 11(2): 555809. DOI:10.19080/JAICM.2021.11.555809.

(77%) and *Pseudomonas aeruginosa* (41%) were the most frequent microorganisms related in this infection. (Supplementary Table 2) Incidence rate of ventilator associated pneumonia was not significantly higher in this group with 13.2 (CI95% 7.24-21.8) per 1000-days-ventilated COVID-19 patients in comparison with 7.1 (CI 1.9-18.3) per 1000-days-ventilated patients in influenza pneumonia cohort (p=0.28). ICU blood stream superinfections were similar in both cohorts (40 vs 47%; p=0.47) with and incidence rate (including secondary bacteraemia) of 25.8 (CI95% 18.1-35.8) in COVID-19 patients and 24,3 (CI95% 15.1-37.2) per 1000-days-at risk in influenza pneumonia cohort (p=0.84). Gram-positive bacteria and Coagulase-negative Staphylococcus were in both cohorts the most frequent bacteria (Supplementary Table 2). The rate of blood stream candidemia was 5 and 14%.

Supplementary Table 2: Aetiology of microbiologically confirmed lower respiratory superinfections in patients with COVID-19 and influenza. All data are expressed as n (%) unless otherwise indicated; COVID-19: Coronavirus disease 2019; ICU: Intensive care unit.

Variables	**Influenza**	**COVID-19**
Lower respiratory superinfections		
Gram-positive bacteria	3 (33.3)	10 (20.8)
Enterococcus spp	3 (33.3)	8 (16.7)
Staphylococcus aureus	0 (0)	2 (4.2)
Gram-negative bacteria	6 (66.7)	37 (77.1)
Escherichia coli	0 (0)	6 12.5)
Pseudomonas aeruginosa	4 (44.4)	20 (41.7)
Klebsiella oxytoca	0 (0)	1 (2.1)
Klebsiella pneumoniae	1 (11.1)	3 (6.3)
Proteus mirabilis	0 (0)	1 (2.1)
Stenotrophomonas maltophilia	0 (0)	4 (8.3)
Achromobacter xyloxidans	0 (0)	1 (2.1)
Delftia acidovorans	1 (11.1)	1 (2.1)
Fungi	0 (0)	1 (2.1)
Aspergillus spp	0 (0)	1 (2.1)
ICU Blood stream superinfections		
Gram-positive bacteria	12 (57.1)	25 (69.4)
Enterococcus faecalis	0 (0)	5 (13.9)
Enterococcus faecium	2 (9.5)	4 (11.1)
Coagulase-negative Staphylococcus	9(42.2)	16 (44.4)
Streptococcus anginosus	1 (4.8)	1 (0)
Gram-negative bacteria	6 (28.6)	9 (25)
Escherichia coli	0 (0)	1 (2.8)
Pseudomonas aeruginosa	5 (23.8)	7 (19.4)
Klebsiella oxytoca	0 (0)	1 (2.8)
Klebsiella pneumoniae	1 (4.8)	0 (0)
Fungi	3 (14.3)	2 (5.6)
Candida spp	3 (14.3)	2 (5.6)
Source of blood stream infection		
Unknown source	12 (57.1)	12 (33.3)
Catheter related	5 (23.8)	16 (44.3)
Respiratory	2 (9.5)	5 (13.9)
Others	2 (9.5)	3 (7.7)

How to cite this article: Rafael L M, Jorge R-G P, Sheila C M, Cristina L M, María Rojas-A. Bacterial and Fungal Infections in Critically Ill Patients with Covid-19 Pneumonia: An Observational Study. J Anest & Inten care med. 2021; 11(2): 555809. DOI:10.19080/JAICM.2021.11.555809.

Variables related with bacterial and/or fungal superinfection in critical care patients with COVID-19

The crude and adjusted analysis of the association between selected variables and superinfection in COVID-19 patients is shown in Table 3. In the crude analysis, variables statically significant related to superinfection were the severity of the illness at admission, SOFA score (Odds Ratio (OR) 1.48; CI 1.13-1.95; p=0.005), interleukin 6 (OR 3.33; CI 1.03-10.74; p=0.04) and days of invasive mechanical ventilation (OR 1.11; CI 1.03-1.20; p=0.005). In adjusted analysis, days of invasive mechanical ventilation (OR 1.08; CI 1.01-1.16; p=0.04) and SOFA score at admission (OR 1.48; CI 1.13-1.95; p=0.04) were statistically associated with superinfection. Treatments with tociluzumab (OR 1.21; CI 0.36-4.10; p=0.75) or corticosteroids (OR 0.49; CI 0.14-1.67; p=0.25) failed to show any relation in the crude and adjusted analysis.

Table 3: Crude and adjusted analysis of the variables associated to bacterial and/or fungal superinfections and patients with COVID-19 admitted in ICU. All data are expressed as n (%) unless otherwise indicated; COVID-19: Coronavirus disease 2019; ICU: Intensive care unit; OR Odds ratio; CI: Confidence interval; IQR: interquartile range; APACHE II score: Acute physiology and chronic health evaluation II; SOFA score: Sequential Organ Failure Assessment.

Variables	Global cohort		Crude analysis		Adjusted analysis 1		Adjusted analysis 2		Adjusted analysis 3	
	Patients without superinfections[a]	Patients with superinfections[b]	OR (CI 95%)	p	OR (CI 95%)	p	OR (CI 95%)	p	OR (CI 95%)	p
Age, median (IQR)	67 (52-74)	63 (53-70)	0.98 (0.94-1.02)	0.44						
Diabetes mellitus	8 (25.8)	8 (29.6)	1.21 (0.38-3.83)	0.74						
Charlson co-morbidity index, median (IQR)	2 (1-4)	3 (2-4)	1.15 (0.85-1.56)	0.34						
Lymphocytes at admission	14 (45.2)	18 (66.7)	0.99 (0.99-1.001)	0.29						
D Dimer > 1500 ng/mL at admission	9 (29)	10 (37)	1.43 (0.47-4.32)	0.51						
IL 6 > 40 pg/mL (n=35), median (IQR)	6 (19.4)	12 (44.4)	3.33 (1.03-10.74)	**0.04**						
APACHE II score, median (IQR)	12 (8-15)	15 (7-18)	1.04 (0.96-1.13)	0.25						
SOFA score at admission, median (IQR)	3 (3-5)	6 (4-7)	1.48 (1.13-1.95)	**0.005**	1.33 (1.01-1.77)	**0.04**				
Days of invasive mechanical ventilation, median (IQR)[c]	0 (0-6)	9 (5-17)	1.11 (1.03-1.20)	**0.005**	1.08 (1.01-1.16)	**0.04**	1.11 (1.03-1.20)	**0.005**	1.12 (1.04-1.21)	**0.003**
Tociluzumab[c]	9 (29)	8 (29.6)	1.02 (0.33-3.19)	0.96			1.21 (0.36-4.10)	0.75		
Corticosteroids[c]	14 (45.2)	11 (40.7)	0.83 (0.29-2.37)	0.73					0.49 (0.14-1.67)	0.25
Non-specific immunoglobulin[c]	5 (16.1)	6 (22.2)	1.48 (0.39-5.55)	0.55						

[a] Patients without bacterial and/or fungal microbiologically confirmed superinfection in ICU.

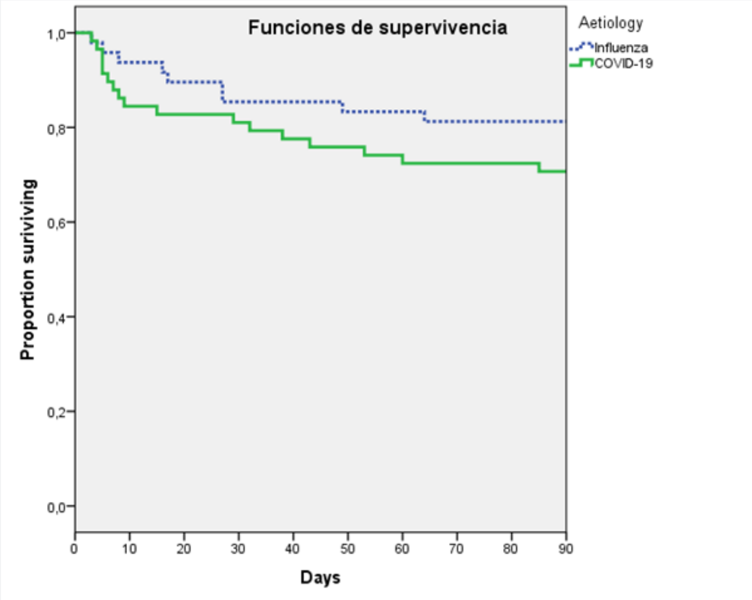
[b] Patients with bacterial and/or fungal microbiologically confirmed superinfection in ICU.

[c] Treatment or support before superinfection episode.

Prognosis in COVID19 patients: Impact of superinfection

Patients with COVID-19 showed higher overall mortality 90 day but there were not statistical differences in comparison with patients with influenza infections (29 vs 18%; p=0.20). Eleven patients died in treatment with active infection in COVID-19 cohort. Survival analysis of 90-day overall mortality did not show differences between COVID-19 and influenza pneumonia (p=0.20). (Supplementary Figure 1)

How to cite this article: Rafael L M, Jorge R-G P, Sheila C M, Cristina L M, María Rojas-A. Bacterial and Fungal Infections in Critically Ill Patients with Covid-19 Pneumonia: An Observational Study. J Anest & Inten care med. 2021; 11(2): 555809. DOI:10.19080/JAICM.2021.11.555809.



Supplementary Figure 1: Survival function of overall moratlity 90 day in patients with COVID-19 and influenza. Log rank test: 0.207.

Table 4 shows that variables that were related in crude analysis with overall mortality at 90 days were severity of illness, APACHE II score (HR 1.07; CI 1.008-1.14; p=0.02), SOFA score at 72 hours (HR 1.19; CI 1.04-1,37; p=0.01) and superinfection (HR 4.76; CI 1.63-13.91; p=0.004) when was explored as time-dependent variable.

Table 4: Crude analysis of the variables associated to overall mortality at 90 day in patients with COVID-19 admitted in ICU. All data are expressed as n (%) unless otherwise indicated; COVID-19: Coronavirus disease 2019; ICU: Intensive care unit; OR Odds ratio; CI: Confidence interval; IQR: interquartile range; APACHE II score: Acute physiology and chronic health evaluation II; SOFA score: Sequential Organ Failure Assessment.

Variables	Crude analysis	
	HR (CI 95%)	p
Age, median (IQR)	1.02 (0.98-1.07)	0.23
Diabetes mellitus	2.05 (0.78-5.39)	0.14
Obesity	2.19 (0.84-5.69)	0.1
Charlson comorbidity index, median (IQR)	1.27 (0.98-1.64)	0.06
APACHE II score, median (IQR)	1.07 (1.008-1.14)	**0.02**
SOFA score at 72 hours, median (IQR)	1.19 (1.04-1.37)	**0.01**
Invasive mechanical ventilation	4.16 (0.95-18.24)	0.05
Microbiologically confirmed coinfection	1.46 (0.33-6.40)	0.61
Microbiologically confirmed superinfection[a]	4.76 (1.63-13.91)	**0.004**
Tociluzumab	1.01 (0.35-2.89)	0.97
Corticosteroids	1.53 (0.59-3.96)	0.38

[a]Microbiologically confirmed superinfection was included as a time-dependent covariate.

Discussion

We found that in our cohort of critically ill patients with COVID19 pneumonia, patients experienced low coinfection rate (3%) and high incidence of superinfections (46%). Gram negative infections were significantly higher in comparison with those with influenza pneumonia and lower respiratory infections and *Pseudomonas aeruginosa* were the most frequent source and microorganism related.

Microbiologically confirmed respiratory co-infection of COVID-19 patients of our cohort was significantly lower (3%) than

How to cite this article: Rafael L M, Jorge R-G P, Sheila C M, Cristina L M, María Rojas-A. Bacterial and Fungal Infections in Critically Ill Patients with Covid-19 Pneumonia: An Observational Study. J Anest & Inten care med. 2021; 11(2): 555809. DOI:10.19080/JAICM.2021.11.555809.

patients with influenza pneumonia. Similar findings are noted in other studies, so we could suggest that coinfection is unlikely to be common in patients with COVID-19 upon admission to the ICU [11,12]. On the other hand, we are aware that coinfection could be influenced by seasonal factors and it is unknown if these results could change during other periods. Considering the low coinfection rate, empiric antibiotic treatment should be closely revaluated once bacterial and/or fungal bacterial infections has been ruled out to reduce resistant emergence. We found 3 cases at admission of bloodstream infections by Coagulase-negative Staphylococcus; in the context of fulfil bloodstream infection criteria and clinical systemic inflammatory response they were treated. We could not rule out that those results as contaminations.

The published incidence of superinfections in critical care patients with COVID-19 is highly variable. While some reports show low incidence of lower respiratory infections less than 30%, other studies point to SARS-CoV-2 as an independent risk factor and to increase its´ incidence (>50%) [5,6,13,14]. In our study, the incidence of lower respiratory infection is elevated (42% of lower respiratory infection and 21% of ventilator associated pneumonia) and higher than those patient with influenza pneumonia. Even though we found lower incidence rate of ICU bloodstream infection than other reports like Giacobbe *et al.* [15] study (47 vs 28 episodes per 1000 patients-days at risk in our COVID-cohort), and was not significantly higher than influenza patients, still remains high with up to 20% incidence of catheter related blood stream infections [15].

Some reasons that could explain this high variability between studies may be: different pressure of ICU admissions and its´ impact in quality care, publication bias (tend to report good results), different severity of illness and patients characteristics, the effect of mortalities differences in superinfection incidence (competitive risk bias) and exclusion in some studies of patients that are still admitted to hospital or ICU at the end of the studies follow up, which could corresponded with long term patients with high rate of complications [12-14,16].

Critical COVID-19 patients are exposed to important risk factors for superinfection that can explain the high rate established like advanced age, underlying systemic diseases, acute respiratory distress syndrome, mechanical ventilation, catheters and prolonged hospital and ICU stays [17,18]. In our case we found that the severity of presentation (SOFA score at admission) interleukin 6 and supportive treatment (days of invasive mechanical ventilation) showed to be more important factors associated with superinfections in that of critical care patients with COVID-19. Even though we were unable to assess if immunomodulatory treatment was related, the study is not powdered enough to test this hypothesis; published data shows controversial findings related corticosteroids and tociluzumab: the balance of the result of theatrically reduction of supports with immunomodulatory treatment (e.g. mechanical ventilation and prognosis) and the increase of the exposure to superinfections is still unknown [9]. Some studies point to SARS-COV-2 as an independent risk factor for exposure to lower respiratory and blood stream infections. Suggested explanations for this hypothesis are endothelial dysfunction, widespread thrombosis, histologic findings of acute fibrinous and organizing pneumonia and gut biota disruption [6,9].

In some papers increasing *Pseudomonas aeruginosa* superinfections in COVID-19 patients were noted [6,19]. In our case, *Pseudomonas aeruginosa* was too the main pathogen isolated (27%). Future studies will assess if COVID-19 clinical evolution and its´ histopathology lung damage/ADRS may predispose to these kinds of infections; despite this, we found that gram negative infections were significantly higher in COVID-19 patients in comparison with influenza pneumonia, local flora may play an important role in this issue.

In our case we can show that superinfection was independently associated with overall mortality at 90 days in crude analysis, but future studies will clear the complex relationship between severity of illness, superinfection, treatments, and mortality.

Our study has obvious limitations. Its´ observational design is exposed to certain biases by its nature, even though we tried to minimize them by applying strict definitions and following STROBE recommendations for observational studies. We are aware of the important limitation of sample size; that is important in the precision of our estimate, so it must be taken with caution. Epidemiology may be influenced by local flora. This cohort of critical care patients with COVID-19 were admitted to ICU before RECOVERY trial results, so more than one half of patients did not receive corticosteroids, it can impact in the future patients and superinfections events [20]. But considering the context of the high variable data reports, these results in this critical care cohort specifically focused on bacterial and/or fungal infection in COVID-19 patients may be useful discussing issues such as the co-infection and superinfection, it´s impact and epidemiology and treatment approach.

Conclusion

The incidence of coinfections is low in critically ill patients with COVID-19 pneumonia and significantly less frequent than in patients with influenza pneumonia. Empirical treatment at admission should be closely revaluated. Incidence of superinfection is high, if is suspected, empirical antipseudomonal treatment should be considered specially in lower respiratory infections, considering local flora. Severity of illness and supportive treatments seems to be the *clearer* factors related to superinfections.

Author's contribution: All the authors equally contributed to the idea presented in the manuscript. The authors read and approved the final manuscript.

Financial support: Collaboration of authors who are members of the Spanish Network for Research in Infectious Diseases (REIPI RD16/0016/0008/ REIPI RD16/0016/0001). No other financial support.

Competing interests: JTC has served as scientific advisor for Pfizer, MSD, Menarinia nd Shionohy; and has received payment for lectures including service on speakers' bureaus and for development of educational presentations for Pfizer and MSD.

References

1. Clancy CJ, Nguyen MH. COVID-19, superinfections and antimicrobial development: What can we expect?. Clin Infect Dis. 2020;ciaa524. doi:10.1093/cid/ciaa524. Online ahead of print.
2. Ferrer R. COVID-19 Pandemic: the greatest challenge in the history of critical care. Medicina Intensiva. 2020; 44(6): 323–324.
3. Betônico GN, Azevedo LC. Approaching COVID-19-Bedside strategies for intensive care. Eur J Clin Invest. 2020; 50(7):e13310.
4. Rawson TM, Moore LSP, Zhu N, Ranganathan N, Skolimowska K, Gilchrist M, Satta G, Cooke G, Holmes A. Bacterial and fungal co-infection in individuals with coronavirus: A rapid review to support COVID-19 antimicrobial prescribing. Clin Infect Dis. 2020; 2;ciaa530. doi: 10.1093/cid/ciaa530. Online ahead of print.
5. Yang X, Yu Y, Xu J, Shu H, Xia J,et al. Clinical course and outcomes of critically ill patients with SARS-CoV-2 pneumonia in Wuhan, China: a single-centered, retrospective, observational study. Lancet Respir Med. 2020; 8(5):475-481. Epub 2020 Feb 24. Erratum in: Lancet Respir Med. 2020 Apr;8(4):e26.
6. Rouzé A, Martin-Loeches I, Povoa P, Makris D, Artigas A, Bouchereau M et al. Relationship between SARS-CoV-2 infection and the incidence of ventilator-associated lower respiratory tract infections: a European multicenter cohort study. Intensive Care Med. 2021. 3;1-11. Online ahead of print.
7. von Elm E, Altman DG , Egger M , et al. Initiative The strengthening the reporting of observational studies. J Clin Epidemio. 2008; 61(4):344-9.
8. Horan TC, Andrus M, Dudeck MA. CDC/NHSN Surveillance Definition of Health Care-Associated Infection and Criteria for Specific Types of Infections in the Acute Care Setting. Am J Infect Control. 2008; 36(5):309-32.
9. Buetti N, Ruckly S, de Montmollin E, Reignier J, Terzi N, Cohen Y, Shiami S, Dupuis C, Timsit JF. COVID-19 increased the risk of ICU-acquired bloodstream infections: a case-cohort study from the multicentric OUTCOMEREA network. Intensive Care Med. 2021; 47(2):180-187.
10. Beyersmann J, Gastmeier P, Wolkewitz M, Schumacher M. J. An easy mathematical proof showed that time-dependent bias inevitably leads to biased effect estimation. Clin Epidemiol. 2008;61(12):1216-1221.
11. Richardson S, Hirsch JS, Narasimhan M, et al. Presenting Characteristics, Comorbidities, and Outcomes Among 5700 Patients Hospitalized With COVID-19 in the New York City Area. JAMA. 2020 Apr 22;323(20):2052-2059. Online ahead of print.
12. Garcia-Vidal C, Sanjuan G, Moreno-García E, Puerta-Alcalde P, Garcia-Pouton N et al. Incidence of co-infections and superinfections in hospitalized patients with COVID-19: a retrospective cohort study. COVID-19 Researchers Group. Clin Microbiol Infect. 2021;27(1):83-88.
13. Ramírez P, Gordón M, Martín-Cerezuela M, Villarreal E, Sancho E, Padrós E, Frasquet J, Leyva G, Molina I, Barrios M, Gimeno S, Castellanos Á. Acute respiratory distress syndrome due to COVID-19. Clinical and prognostic features from a medical Critical Care Unit in Valencia, Spain. Med Intensiva. 2020;11;S0210-5691(20)30245-X.
14. Ferrando C, Mellado-Artigas R, Gea A, Arruti E, Aldecoa C,et al, de la Red de UCI Española para COVID-19. Patient characteristics, clinical course and factors associated to ICU mortality in critically ill patients infected with SARS-CoV-2 in Spain: A prospective, cohort, multicentre study. Rev Esp Anestesiol Reanim. 2020; 67(8):425-437.
15. Giacobbe DR, Battaglini D, Ball L, Brunetti I, Bruzzone B, Codda G, Crea F, De Maria A, Dentone C, Di Biagio A, Icardi G, Magnasco L, Marchese A, Mikulska M, Orsi A, Patroniti N, Robba C, Signori A, Taramasso L, Vena A, Pelosi P, Bassetti M. Bloodstream infections in critically ill patients with COVID-19. Eur J Clin Invest. 2020;50(10):e13319.
16. Bassetti M, Kollef MH, Timsit JF. Bacterial and fungal superinfections in critically ill patients with COVID-19. Intensive Care Med. 2020;46(11):2071-2074.
17. Rynda-Apple A, Robinson KM, Alcorn JF. Influenza and bacterial superinfection: illuminating the immunologic mechanisms of disease. Infect Immun 2015;83(10):3764–70.
18. Bauer TT, Ewig S, Rodloff AC, Müller EE. Acute Respiratory Distress Syndrome and Pneumonia: A Comprehensive Review of Clinical Data. Clin Infect Dis. 2006; 15;43(6):748-56.
19. Arentz M, Yim E, Klaff L, et al. Characteristics and Outcomes of 21 Critically Ill Patients With COVID-19 in Washington State. JAMA. 2020;323(16):1612-1614.
20. RECOVERY Collaborative Group, Horby P, Lim WS, Emberson JR, Mafham M, Bell JL, Linsell L, Staplin N, Brightling C, Ustianowski A, Elmahi E, Prudon B, Green C, Felton T, Chadwick D, Rege K, Fegan C, Chappell LC, Faust SN, Jaki T, Jeffery K, Montgomery A, R. Dexamethasone in Hospitalized Patients with Covid-19 - Preliminary Report. N Engl J Med. 2020; 17:NEJMoa2021436.

How to cite this article: Rafael L M, Jorge R-G P, Sheila C M, Cristina L M, María Rojas-A. Bacterial and Fungal Infections in Critically Ill Patients with Covid-19 Pneumonia: An Observational Study. J Anest & Inten care med. 2021; 11(2): 555809. DOI:10.19080/JAICM.2021.11.555809.