Detection of bacteria via multiplex PCR in respiratory samples of critically ill COVID-19 patients with suspected HAP/VAP in the ICU

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Summary
Background Critically ill Coronavirus disease 2019 (COVID-19) patients have high rates of bacterial superinfection. Multiplex polymerase chain reaction panels may be able to provide useful information about the incidence and spectrum of bacteria causing superinfections.

Methods In this retrospective observational study we included all COVID-19 positive patients admitted to our intensive care unit with suspected hospital-acquired pneumonia/ventilator-associated pneumonia (HAP/VAP) in whom the BioFire® Pneumonia Panel (PP) was performed from tracheal aspirate or bronchoalveolar lavage fluid for diagnostic purposes. The aim of our study was to analyze the spectrum of pathogens detected with the PP.

Results In this study 60 patients with a median age of 62.5 years were included. Suspected VAP was the most frequent (48/60, 80%) indication for performing the PP. Tracheal aspirate was the predominant sample type (50/60, 83.3%).

The PP led to a negative, monomicrobial and polymicrobial result in 36.7%, 35% and 28.3% of the patients, respectively. The three most detected bacteria were Staphylococcus aureus (13/60, 21.7%), Klebsiella pneumoniae (12/60, 20%) and Haemophilus influenzae (9/60, 15%). Neither atypical bacteria nor resistance genes were detected.

Microbiological culture of respiratory specimens was performed in 36 (60%) patients concomitantly. The PP and microbiological culture yielded a non-concordant, partial concordant and completely concordant result in 13.9% (5/36), 30.6% (11/36) and 55.6% (20/36) of the analyzed samples, respectively.

Conclusion In critically ill COVID-19 patients with suspected HAP/VAP results of the PP and microbiological culture methods were largely consistent. In our cohort, S. aureus and K. pneumoniae were the most frequently detected organisms. A higher diagnostic yield may be achieved if both methods are combined.

Keywords Superinfection · Mortality · Biofire · Pneumonia panel · Intensive care unit

Introduction

The clinical spectrum of coronavirus disease 2019 (COVID-19) ranges from completely asymptomatic or mild manifestations to severe and life-threatening forms requiring treatment in an intensive care unit [1–5]. Early studies have suggested that approximately 7% of patients are affected by bacterial co/superinfections [6, 7]. One meta-analysis showed that 3.5% of COVID-19 patients had a bacterial coinfection on admission and 14.3% developed a bacterial superinfection during hospital stay but 72% of all patients received empirical antibiotic treatment, mainly with third generation cephalosporins and respiratory fluoroquinolones (e.g. levofloxacin) [8]. In contrast, rates of bacterial superinfections are higher in influenza positive patients and range from 20% to 30% [9].

According to observational data, COVID-19 patients treated in the intensive care unit (ICU) have higher rates of bacterial superinfections than patients
on normal wards [8, 10]. Interestingly, the ventilator-associated pneumonia rates are higher in COVID-19 patients ranging from 29% to 57% [11–14] compared to non-COVID-19 patients, where the incidence is approximately 10% (range 5–40%, depending on the underlying population) [11, 15, 16].

While multiplex polymerase chain reaction (PCR) based panels from cerebrospinal fluid on top of standard diagnostic procedures for suspected meningitis/encephalitis have been shown to have excellent diagnostic accuracy in two meta-analyses [17, 18], data for respiratory tract infections are conflicting, with some studies indicating a decrease in length of stay and antibiotic prescriptions [19, 20], while others failed to demonstrate any impact [21–23].

The aim of our retrospective, observational study was to analyze the spectrum of pathogens detected with multiplex PCR and culture from respiratory samples in critically ill COVID-19 patients with suspected hospital-acquired pneumonia (HAP) or ventilator-associated pneumonia (VAP).

Methods

Study design and population

This retrospective observational study took place at the Department for Infectious Diseases, Klinik Favoriten, in Vienna, Austria. All patients with PCR confirmed COVID-19 infections treated in our ICU between March and October 2020 with suspected HAP or VAP in whom the BioFire® Pneumonia Panel (PP) (bioMérieux SA RCS, Lyon, France) was performed were included in the study. The PP was carried out by trained medical staff at our point-of-care laboratory from respiratory samples (tracheal aspirate or bronchoalveolar lavage fluid) collected immediately before the test. The decision to utilize the PP was based on clinical grounds (e.g., deterioration, fever, rise in inflammatory markers, purulent secretion, new consolidations on chest X-ray) by the treating physicians. The PP is a multiplex PCR designed to detect the most important pathogens of viral (adenovirus, coronaviruses, human metapneumovirus, human rhinovirus/enterovirus, influenza A/B, parainfluenza virus, respiratory syncytial virus) and bacterial (Acinetobacter calcoaceticus-baumannii, Enterobacter cloacae complex, Escherichia coli, Haemophilus influenzae, Klebsiella aerogenes, Klebsiella oxytoca, Klebsiella pneumoniae group, Moraxella catarrhalis, Proteus spp, Pseudomonas aeruginosa, Serratia marcescens, Staphylococcus aureus, Streptococcus agalactiae, Streptococcus pneumoniae, Streptococcus pyogenes, Chlamydia pneumoniae, Legionella pneumophila, Mycoplasma pneumoniae) pneumonia within less than 2 h. Additionally, it is capable of detecting the most common antibiotic resistance genes (mecA/C and MREJ, CTX-M, KPC, NDM, VIM, IMP, OXA-48-like) [24].

If a respiratory sample for conventional microbiological culture was obtained concomitantly, we compared the results with the PP and classified them as completely concordant (all detected pathogens matched in both tests or no pathogens was found), partially concordant (both tests detected the same pathogen, but an additional pathogen was detected in either the PP or culture) or non-concordant (pathogen detected in either PP or culture differed from each other).

For example, if S. aureus plus H. influenzae were detected as the only pathogens in PP and culture this was classified as completely concordant. If the PP result was S. aureus and H. influenzae but culture grew only S. aureus this was classified as partially concordant. If the PP result was P. aeruginosa and the culture result was negative or the PP result was H. influenzae and the culture result was E. coli, this was classified as non-concordant. Clinically irrelevant organisms that are generally regarded as contaminants or typical colonizers of the respiratory tract were excluded from the analysis.

Definition of variables

Hospital-acquired pneumonia and VAP were defined as pneumonia occurring ≥48 h after hospital admission and ≥48 h after intubation, respectively. A combination of the following clinical criteria led to the suspicion of HAP/VAP: new onset of fever, increased purulent sputum, worsening of respiratory function, and detection of a new pulmonary consolidations [25].

Statistical analysis and data collection

Data were collected from patient medical records, entered in a MS Excel sheet (Microsoft, Redmond, WA, USA) and anonymized before statistical analysis. All analyses were made with SPSS 25 (IBM, Armonk, NY, USA) for Mac OS (Apple, Cupertino, CA, USA). Categorical variables were described by counts and percentage. For metric, non-normally distributed variables the median (Md) and interquartile range (IQR) were used. Significance tests for categorical variables were made via cross-tables and χ²-tests or Fisher's exact test where applicable. A two-sided alpha <0.05 was considered statistically significant.

The study was approved by the ethics committee of the City of Vienna (EK 20-079). All methods were carried out in accordance with the ethical principles of the declaration of Helsinki.

Results

Demographics

A total of 60 patients with a median age of 62.5 years (IQR 52–71.75 years) admitted to the ICU between March and October 2020 were included in the study.
The majority of the patients were male (48/60, 80%). Hypertension (66.8%), type 2 diabetes mellitus (30%) and coronary heart disease (20%) were the most common comorbidities. Time from symptom onset to hospitalization and ICU admission was 5 days (IQR 3–8 days) and 7 days (IQR 5–10 days) respectively.

Of the patients 22 (36.7%) died during the hospital stay. Median length of stay in the ICU of survivors was 24.5 days (IQR 18–30.75 days) and total hospital length of stay of survivors was 41 days (IQR 30–62.5 days). For details see Table 1.

**Results of the PP and microbiological culture**

Of the PPs 80% were performed in patients with suspected VAP and in 20% with suspected HAP. Tracheal aspirates and bronchoalveolar lavages (BAL) were analyzed in 83% and 17% of the patients, respectively. The PP was performed on average 7.5 days [3–10, 17, 18] after ICU admission. Most patients (73%) received an antibiotic at the time when the PP was performed (Table 1).

### Table 1 Patient characteristics

|                       | All patients |          |          |
|-----------------------|--------------|----------|----------|
| Sex (male)            | 48/60 (80%)  |          |          |
| Age in years (median, IQR) | 62.5 (52–71.75) |          |          |
| BMI (median, IQR)     | 29 (26–36.75) |          |          |
| Hypertension          | 40/60 (66.7%)|          |          |
| Diabetes mellitus, type 2 | 18/60 (30%)  |          |          |
| Coronary heart disease| 12/60 (20%)  |          |          |
| Chronic kidney disease| 6/60 (10%)   |          |          |
| Time from symptom onset before hospital admission (n = 58) in days (median, IQR) | 5 days (3–8) |          |          |
| Time from symptom onset before ICU admission (n = 58) in days (median, IQR) | 7 days (5–10) |          |          |

### Table 2 Specific results from the pneumonia panel and microbiological culture

|                       | Pneumonia panel |          | Microbiological culture |
|-----------------------|-----------------|----------|-------------------------|
|                       | N = 60          |          | N = 36                  |
| Staphylococcus aureus | 13 (21.7%)      | 10 (27.8%)|                          |
| Klebsiella pneumoniae| 12 (20%)        | 9 (25%)  |                          |
| Haemophilus influenzae| 9 (15%)        | 2 (5.6%) |                          |
| Echeria coli          | 5 (8.3%)        | 2 (5.6%) |                          |
| Streptococcus pneumonia| 5 (8.3%)    | 0 (0%)   |                          |
| Pseudomonas aeruginosa| 3 (5%)         | 3 (8.3%) |                          |
| Serratia marcesens    | 3 (5%)          | 2 (5.6%) |                          |
| Klebsiella aerogenes  | 3 (5%)          | 3 (8.3%) |                          |
| Streptococcus agalactiae| 2 (3.3%)  | 2 (5.6%) |                          |
| Klebsiella oxytoca    | 2 (3.3%)        | 1 (2.8%) |                          |
| Acinetobacter baumannii| 2 (3.3%)    | 0 (0%)   |                          |
| Enterobacter cloacae  | 2 (3.3%)        | 2 (5.6%) |                          |
| Proteus spp           | 2 (3.3%)        | 0 (0%)   |                          |
| Other bacteria1       | 0 (0%)          | 10 (27.7%)|                          |

1Other bacteria detected: Burkholderia cepacia (1), Citrobacter koserii (1), Enterococcus faecalis (1), Roseateles aquatilis (1), Raoultella planticola (1), Serratia liquefaciens (2), Staphylococcus lugdunensis (1), Viridans group streptococci (1)

### Table 3 General results PP and microbiological culture

|                       | All patients | On antibiotics when tested | p-value |
|-----------------------|--------------|----------------------------|---------|
|                       | N = 60       | No                        | Yes     |         |
| Results of PP (n = 60) |              |                            |         |
| Negative              | 22/60 (36.7%)| 3/16 (18.8%)              | 19/44 (43.2%)| 0.151   |
| Monomicrobial         | 21/60 (35%)  | 6/16 (37.5%)              | 15/44 (34.1%)|         |
| Polymicrobial         | 17/60 (28.3%)| 7/16 (43.8%)              | 10/44 (22.7%)|         |
| Results from culture (n = 36) |         |                            |         |
| Negative              | 12/36 (33.3%)| 2/0 (22.2%)               | 10/27 (37.1%)| 0.710   |
| Monomicrobial         | 15/36 (41.7%)| 4/0 (44.4%)               | 11/27 (40.7%)|         |
| Polymicrobial         | 9/36 (25%)   | 3/0 (33.3%)               | 6/27 (22.2%)|         |
| Comparison PP and culture (n = 36) |         |                            |         |
| Non-concordant        | 5/36 (13.9%) | –                         | –       |         |
| Partially concordant  | 11/36 (30.6%)| –                         | –       |         |
| Completely concordant | 20/36 (55.6%)| –                         | –       |         |

**PP pneumonia panel**
Overall, the PP led to a negative, monomicrobial and polymicrobial result in 36.7%, 35% and 28.3% of the patients, respectively. The five most commonly detected pathogens were *S. aureus* (13/60, 21.7%), *K. pneumoniae* (12/60, 20%), *H. influenzae* (9/60, 15%), *E. coli* (5/60, 8.3%) and *S. pneumoniae* (5/60, 8.3%) (see Table 2).

No resistance genes, and no viruses or atypical bacteria were detected.

In 36 patients a microbiological culture of a respiratory specimen was performed concomitantly with the PP. The microbiological culture led to a negative, monomicrobial and polymicrobial result in 33.3%, 41.7% and 25% of the patients, respectively. The four most commonly detected pathogens were *S. aureus* (10/36, 27.8%), *K. pneumoniae* (9/36, 25%), *K. aerogenes* (3/36, 8.3%) and *P. aeruginosa* (3/36, 8.3%). Other detected bacteria are listed in Table 2.

Of the patients 75% (27/36) had a positive PP and/or culture result. Antibiotic administration had no statistically significant effect on the results of the PP ($p=0.151$) and culture ($p=0.710$) but patients on antibiotics had a numerically higher rate of negative PP and culture results, as shown in Table 3.

The PP and microbiological culture yielded a non-concordant, partial concordant and completely concordant result in 13.9% (5/36), 30.6% (11/36) and 55.6% (20/36) of the analyzed samples, respectively. For details see Table 3.

### Discussion

In patients with COVID-19 treated in our ICU with suspected HAP/VAP, the rate of positive PP results was high. Monomicrobial, polymicrobial and negative results were found in approximately 30% each. The most frequently detected bacteria were *S. aureus, K. pneumoniae* and *H. influenzae*. Microbiological culture led to similar results in our study population, where *S. aureus* and *K. pneumoniae* have been detected most frequently.

In other studies with COVID-19 patients *S. aureus* und *S. pneumoniae* have been identified as the most important pathogens in community-acquired pulmonary superinfections, while in hospital-acquired superinfections *S. aureus, H. influenzae, K. pneumoniae* and, on some occasions, non-fermenting bacteria such as *P. aeruginosa* and *A. baumannii* predominated [6–8, 10, 26, 27]. In our study non-fermenting bacteria were only detected occasionally. One study found a high rate of atypical bacteria in COVID-19 co-infections but these diagnoses were based on serological tests, which can generally not be regarded as a reliable tool [7]. In contrast, we detected neither any atypical bacteria nor viruses other than severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). Empirical antibiotic coverage for atypical bacteria in COVID-19 is not recommended in the guidelines [28].

A major advantage of multiplex PCRs and their use in point-of-care laboratories is the fast turn-around time permitting the treating physicians to establish a targeted anti-infective therapy more rapidly than with conventional methods. The PP delivers results of the most common bacteria and resistance genes within less than 2 h with very little hands-on time; however, to interpret the results properly and to draw the correct conclusions expert knowledge on infectious diseases as well as antimicrobial therapy is necessary, especially when it comes to interpreting polymicrobial results. Furthermore, interpretation is complicated if results from the PP and microbiological culture differ.

We were able to demonstrate that the results from the PP and concomitantly microbiological culture were completely concordant in approximately 55.6% and partially concordant in 30.6%, emphasizing the usefulness of point-of-care multiplex PCRs additionally to microbiological techniques, especially when classical microbiological culture methods are not available due to laboratory opening hours or delayed transportation time and regarding slower results from culture. Recently a study demonstrated high concordance between conventional microbiological culture and the PP in non-COVID-19 patients admitted to hospital for lower respiratory tract infections. Interestingly, the PP showed polymicrobial results in almost half of the patients, with *S. aureus* and *H. influenzae* being the most detected bacteria via the PP [29]. While the high incidence of *S. aureus* in the PP was confirmed by culture in our study, *H. influenzae* was more frequently found in the PP alone. This raises the question if the prevalence of *H. influenzae* may be overestimated by using PCR-based methods.

The detection of bacteria via multiplex PCR based methods may only reflect colonization and not infection on some occasions, which is one of the main shortcomings of such diagnostic tools. Furthermore, despite the potential detection of major resistance genes such methods lack the ability of generating detailed antimicrobial susceptibility profiles. Resistance rates in Austria are low compared to most other countries [30], which might explain why no resistance genes were identified in our patient cohort. Additionally, the rate of empirical antibiotic prescription is lower at our department compared to other hospitals which might contribute to low resistance gene detection in our ICU cohort [8, 31]. The majority of patients are transferred from our normal ward to the ICU and are not directly admitted to the ICU.

The administration of antibiotics had no statistically significant effect on the detection rate of bacteria in the PP and culture but patients on antibiotics had a numerically higher rate of negative results in both groups. While each method on its own detected any bacteria in approximately two thirds of the patients, 75% of the patients had either a positive PP and/or culture result. This supports the additional
use of both diagnostic methods. Very recently, studies have shown the potential of multiplex PCR to improve antibiotic treatment in bacterial pneumonia in non-COVID-19 and COVID-19 patients [32, 33].

Limitations of our study are the small sample size and the single center study design. Furthermore, the decision to perform the PP was not guided by specific criteria but was based on clinical judgement of the treating physician. Moreover, we neither performed a microbiological culture in all patients, nor did we test for fungal infections routinely, despite the increasing number of publications regarding COVID-19-associated pulmonary aspergillosis [34, 35]. We did not have a control group of patients with other viral disease like influenza to compare. The strength of our study is that it reflects common practice in clinical routine. Little is known about the usability of multiplex PCR in the setting of a pandemic. Additionally, our results can give clinicians guidance on which pathogens should be covered empirically in COVID-19 patients at ICUs when diagnostic results are pending or not available. The local epidemiology and resistance rates have to be taken into account.

In summary, in critically ill COVID-19 patients the PP provides fast results with high detection rates and consistent results in comparison to culture. VAP is a very common complication of COVID-19 mechanically ventilated patients, with S. aureus, K. pneumoniae and H. influenzae being the most frequently detected organisms in our cohort. Due to a lack of stringent, evidence-based recommendations, the interpretation of PCR results, especially if they are polymicrobial, remains challenging. While many issues regarding the proper use of multiplex PCRs are yet unresolved, these diagnostic tools may help clinicians to obtain additional information faster in everyday clinical routine and should be implemented when possible. A higher diagnostic yield may be achieved if multiplex PCR based methods are combined with microbiological culture techniques. In our cohort 75% of the patients had either a positive PP and/or culture result, while each method on its own detected any bacteria in only approximately two thirds of the patients.

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Author Contribution Mario Karolyi and Erich Pawelka had the idea of the study. Mario Karolyi wrote the manuscript. Julian Hind, Sebastian Baumgartner, Wolfgang Hoepler, Sara OmId and Tamara Seitz collected the data. Mario Karolyi, Emanuela Friese, Stephanie Neuhold and Marianna Traugott analyzed the data. Christoph Wenisch and Alexander Zoufaly supervised the study.

Declarations

Conflict of interest M. Karolyi, E. Pawelka, J. Hind, S. Baumgartner, E. Friese, W. Hoepler, S. Neuhold, S. OmId, T. Seitz, M.T. Traugott, C. Wenisch and A. Zoufaly declare that they have no competing interests.

Ethical standards This retrospective study was performed after consultation with the institutional ethics committee and in accordance with national legal requirements. The study was approved by the ethics committee of the capital city Vienna. Consent for publications not applicable.

References

1. 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;323(20):2052–9. https://doi.org/10.1001/jama.2020.6775.
2. Serafin RB, Póvoa P, Souza-Dantas V, Kalic AC, Salluh JIF. Clinical course and outcomes of critically ill patients with COVID-19 infection: a systematic review. Clin Microbiol Infect. 2021;27(1):47–54. https://doi.org/10.1016/j.cmi.2020.10.017.
3. Zhou F, Yu T, Du R, et al. Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study. Lancet. 2020;https://doi.org/10.1016/S0140-6736(20)30566-3.
4. Wu Z, McGoogan JM. Characteristics of and important lessons from the Coronavirus disease 2019 (COVID-19) outbreak in China: summary of a report of 72,314 cases from the Chinese center for disease control and prevention. JAMA. 2020;323(13):1239–42. https://doi.org/10.1001/jama.2020.2648.
5. Abate SM, Ahmed Ali S, Mantfardo B, et al. Rate of intensive care unit admission and outcomes among patients with coronavirus: a systematic review and meta-analysis. PLoS One. 2020;15(7):e235653. https://doi.org/10.1371/journal.pone.0235653.
6. Garcia-Vidal C, Sanjuan G, Moreno-Garcia E, et al. Incidence of co-infections and superinfections in hospitalized patients with COVID-19: a retrospective cohort study. Clin Microbiol Infect. 2020;https://doi.org/10.1016/j.cmi.2020.07.041.
7. Lansbury L, Lim B, Baskaran V, Lim WS. Co-infections in people with COVID-19: a systematic review and meta-analysis. J Infect. 2020;81(2):266–75. https://doi.org/10.1016/j.jinf.2020.05.046.
8. Langford BJ, So M, Raybardhan S, et al. Bacterial co-infection and secondary infection in patients with COVID-19: a living rapid review and meta-analysis. Clin Microbiol Infect. 2020;https://doi.org/10.1016/j.cmi.2020.07.016.
9. Klein EY, Monteforte B, Gupta A, et al. The frequency of influenza and bacterial coinfection: a systematic review and meta-analysis. Influenza Other Respir Viruses. 2016;10(5):394–403. https://doi.org/10.1111/irv.12398.
10. Søgaard KK, Baettig V, Osthoff M, et al. Community-acquired and hospital-acquired respiratory tract infection and bloodstream infection in patients hospitalized with COVID-19 pneumonia. J Intensive Care. 2021;9(1):10. https://doi.org/10.1186/s40560-021-00526-y.
11. Maes M, Higginson E, Pereira-Dias J, et al. Ventilator-associated pneumonia in critically ill patients with COVID-19. Crit Care. 2021;25(1):25. https://doi.org/10.1186/s13054-021-03460-5.
12. COVID-ICU Group. Clinical characteristics and day-90 outcomes of 4244 critically ill adults with COVID-19: a prospective cohort study. Intensive Care Med. 2021;47(1):60–73. https://doi.org/10.1007/s00134-020-06294-x.
13. Giacobbe DR, Battaglini D, Enrile EM, et al. Incidence and prognosis of ventilator-associated pneumonia in critically ill patients with COVID-19: a multicenter study. J Clin Med. 2021;10(4):555. https://doi.org/10.3390/jcm10040555.
14. d’Humières C, Patrier J, Lortat-Jacob B, et al. Two original observations concerning bacterial infections in COVID-19 patients hospitalized in intensive care units during the first wave of the epidemic in France. PLoS One. 2021;16(4):e250728. https://doi.org/10.1371/journal.pone.0250728.
15. Papazian L, Klompas M, Luyt CE. Ventilator-associated pneumonia in adults: a narrative review. Intensive Care Med. 2020;46(5):888–906. https://doi.org/10.1007/s00134-020-05980-0.
16. Modi AR, Kovacs CS. Hospital-acquired and ventilator-associated pneumonia: diagnosis, management, and prevention. Cleve Clin J Med. 2020;87(10):633–9. https://doi.org/10.3949/ccjm.87a.19117.
17. Tansarli GS, Chapin KC. Diagnostic test accuracy of the BioFire® FilmArray® meningitis/encephalitis panel: a systematic review and meta-analysis. Clin Microbiol Infect. 2020;26(3):281–90. https://doi.org/10.1016/j.cmi.2019.11.016.
18. Vetter P, Schibler M, Herrmann JL, et al. Accuracy of comprehensive PCR analysis of nasopharyngeal and oropharyngeal swabs for CT-scan-confirmed pneumonia in elderly patients: a randomized controlled trial. Open Forum Infect Dis. 2019;6(12):ofz481. https://doi.org/10.1093/ofid/ofz481.
19. May L, Tairo G, Poltavsky E, et al. Rapid multiplex testing for upper respiratory pathogens in the emergency department: a randomized controlled trial. Open Forum Infect Dis. 2019;6(12):ofz481. https://doi.org/10.1093/ofid/ofz481.
20. Stengchen D, Gu X, Fan G, et al. Evaluation of a molecular point-of-care testing for viral and atypical pathogens on intravenous antibiotic duration in hospitalized adults with lower respiratory tract infection: a randomized clinical trial. Clin Microbiol Infect. 2019;25(11):1415–21. https://doi.org/10.1016/j.cmi.2019.06.012.
21. Busson L, Bartiaux M, Brahim S, et al. Contribution of the filmarray respiratory panel in the management of adult and pediatric patients attending the emergency room during 2015–2016 influenza epidemics: an interventional study. Int J Infect Dis. 2019;83:32–9. https://doi.org/10.1016/j.ijid.2019.03.027.
22. Prendki V, Huttner B, Marti C, et al. Accuracy of comprehensive PCR analysis of nasopharyngeal and oropharyngeal swabs for CT-scan-confirmed pneumonia in elderly patients: a prospective cohort study. Clin Microbiol Infect. 2019;25(9):1114–9. https://doi.org/10.1016/j.cmi.2018.12.037.
23. Saarela E, Tapiainen T, Kauppila J, et al. Impact of multiplex respiratory virus testing on antimicrobial consumption in adults in acute care: a randomized clinical trial. Clin Microbiol Infect. 2020;26(4):506–11. https://doi.org/10.1016/j.cmi.2019.09.013.
24. Biofire. The BioFire® FilmArray® Pneumonia (PN) Panel. 2021. https://www.biofire edx.com/products/the-filmarray-panels/filmarray-pneumonia/. Accessed 5 May 2021.
25. Kalil AC, Metersky ML, Klompas M, et al. Management of adults with hospital-acquired and ventilator-associated pneumonia: 2016 clinical practice guidelines by the infectious diseases society of america and the American thoracic society. Clin Infect Dis. 2017;63(5):e61–e111. https://doi.org/10.1093/cid/ciw353.
26. Contou D, Claudinson A, Pajot O, et al. Bacterial and viral co-infections in patients with severe SARS-CoV-2 pneumonia admitted to a French ICU. Ann Intensive Care. 2020;10(11):119. https://doi.org/10.1186/s13151-020-00736-x.
27. Sharifipour E, Shams S, Esikhmani M, et al. Evaluation of bacterial co-infections of the respiratory tract in COVID-19 patients admitted to ICU. BMC Infect Dis. 2020;20(1):646. https://doi.org/10.1186/s12879-020-05374-z.
28. Sieswerda E, de Boer MGJ, Bonten MMJ, et al. Recommendations for antibacterial therapy in adults with COVID-19—an evidence based guideline. Clin Microbiol Infect. 2021;27(1):61–6. https://doi.org/10.1016/j.cmi.2020.09.041.
29. Webber DM, Wallace MA, Burnham CA, Anderson NW. Evaluation of the biofire filmarray pneumonia panel for detection of viral and bacterial pathogens in lower respiratory tract specimens in the setting of a tertiary care academic medical center. J Clin Microbiol. 2020;58(7):e343–20. https://doi.org/10.1128/JCM.00343-20.
30. Allerberger F, Apfalter P, Bernig L, et al. Resistenzbericht Österreich AURES 2017. Antibiotikaresistenz und Verbrauch antimikrobieller Substanzen in Österreich. Wien: Bundesministerium für Arbeit, Soziales, Gesundheit und Konsumentenschutz; 2017. ISBN978-3-85010-515-6.
31. Karolyi M, Pawelka E, Mader T, et al. Hydroxychloroquine versus lopinavir/ritonavir in severe COVID-19 patients: results from a real-life patient cohort. Wien Klin Wochenschr. 2020. https://doi.org/10.1007/s00508-020-01720-y.
32. Monard C, Pehlivan J, Auger G, et al. Multicenter evaluation of a syndromic rapid multiplex PCR test for early adaptation of antimicrobial therapy in adult patients with pneumonia. Crit Care. 2020;24(1):434. https://doi.org/10.1186/s13054-020-03114-y.
33. Maatouoi N, Chemali L, Patrier J, et al. Impact of rapid multiplex PCR on management of antibiotic therapy in COVID-19-positive patients hospitalized in intensive care unit. Eur J Clin Microbiol Infect Dis. 2021;40(10):2227–34. https://doi.org/10.1007/s10096-021-04213-6.
34. Arastehfar A, Carvalho A, van de Veerdonk FL, et al. COVID-19 associated pulmonary aspergillosis (CAPA) from immunology to treatment. J Fungi (Basel). 2020;6(2):91. https://doi.org/10.3390/jof6020091.
35. Koehler P, Bassetti M, Chakrabarti A, et al. Defining and managing COVID-19-associated pulmonary aspergillosis: the 2020 ECCM/ISHAM consensus criteria for research and clinical guidance. Lancet Infect Dis. 2020. https://doi.org/10.1016/S1473-3099(20)30847-1. 2020;S1473-3099(20)30847-1.