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The value of multiplex PCR in describing the microbial ecology of respiratory infections during the COVID 19 pandemic

Sara Kouara ^{1,2}, Zina Lebbar ^{1,2,*}, Jihad Branya ^{1,2}, Mustapha Mahmoud ^{1,2} and Ghita Yahyaoui ^{1,2}

¹ Department of Microbiology, central medical analysis laboratory, Hassan II university hospital, Fez, Morocco.

² Faculté de Médecine et de Pharmacie Sidi Mohammed Ben Abdellah, Fès, Maroc.

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Abstract

Respiratory infections constitute a frequent reason for consultation and hospitalization and their emergence means that the identification of pathogens is important for the implementation of appropriate treatment for effective patient care. Multiplex PCR is a diagnostic tool developed for the identification of the microbial agent and the detection of resistance mechanisms. The objective of this study was to highlight and identify by multiplex PCR the bacteria and viruses responsible for respiratory infections in patients presenting clinical signs, and in whom biological examinations did not identify germs in cause during the COVID 19 pandemic, to be able to follow the epidemiology and microbial ecology of lower respiratory infections which has undergone changes, due to hygiene rules as well as current measures and methods of controlling this pandemic.

Keywords: Respiratory infections; Multiplex PCR; COVID 19; Microbial ecology.

1 Introduction

The emergence of respiratory infections means that identification of pathogens is important for the implementation of appropriate treatment for effective patient management. Etiological diagnosis is based on classical methods, which have developed and progressed towards new techniques for monitoring the epidemiology and microbial ecology of lower respiratory infections, especially during the COVID 19 pandemic.

Multiplex PCR is a relevant tool for identifying most pathogens responsible for the respiratory pathology observed, and for detecting resistance mechanisms, thus enabling a syndromic approach to respiratory pathology [1]. It is an interesting technique due to its speed and reliability, but many uncertainties remain as to the value of this diagnosis, hence the need for knowledge of the microbial epidemiology of lower respiratory infections.

2 Material and methods

This is a retrospective study spread over 3 years from January 2020 to August 2022, involving 120 samples, received from different hospital wards at the Hassan II University Hospital in Fez.

The aim of this study was to highlight and identify by multiplex PCR the bacteria and viruses responsible for lower respiratory infections in patients presenting clinical signs, and in whom biological examinations did not identify any germs involved during the COVID 19 pandemic, and to compare them with data from the literature.

* Corresponding author: Zina Lebbar

The tool used in our study is the Film Array instrument with the Respiratory panel, which targets 27 clinically relevant pathogens involved in lower respiratory tract infections, as well as 7 antibiotic resistance markers (Table 1) [2].

Table 1 The different germs isolated from the respiratory panel [2]

Bacterium	Atypical Bacteria	Virus	Antibiotic Resistance Genes
<i>Acinetobacter calcoaceticus-baumannii complex</i>	Chlamydia pneumonia	Adénovirus	Methicillin resistance mecA/C et MREJ
<i>Enterobacter cloacae complex</i>	Legionella pneumophila	Coronavirus	Carbapenemases IMP KPC NDM OXA-48-like VIM
<i>Escherichia coli</i>	Mycoplasma pneumonie	Métapneumovirus humain	BLSE CTX-M
<i>Haemophilus influenzae</i>		Human enterovirus/rhinovirus	
<i>Klebsiella aerogenes</i>		Flu virus A	
<i>Klebsiella oxytoca</i>		Flu virus B	
Groupe <i>Klebsiella pneumonie</i>		Coronavirus syndrome respiratory system from the Middle East (MERS CoV)	
<i>Moraxella catarrhalis</i>		Parainfluenza virus	
<i>Proteus spp</i>		Respiratory syncytial virus	
<i>Pseudomonas aeruginosa</i>			
<i>Serratia marcescens</i>			
<i>Staphylococcus aureus</i>			
<i>Streptococcus agalactiae</i>			
<i>Streptococcus pyogenes</i>			
<i>Streptococcus pneumonie</i>			

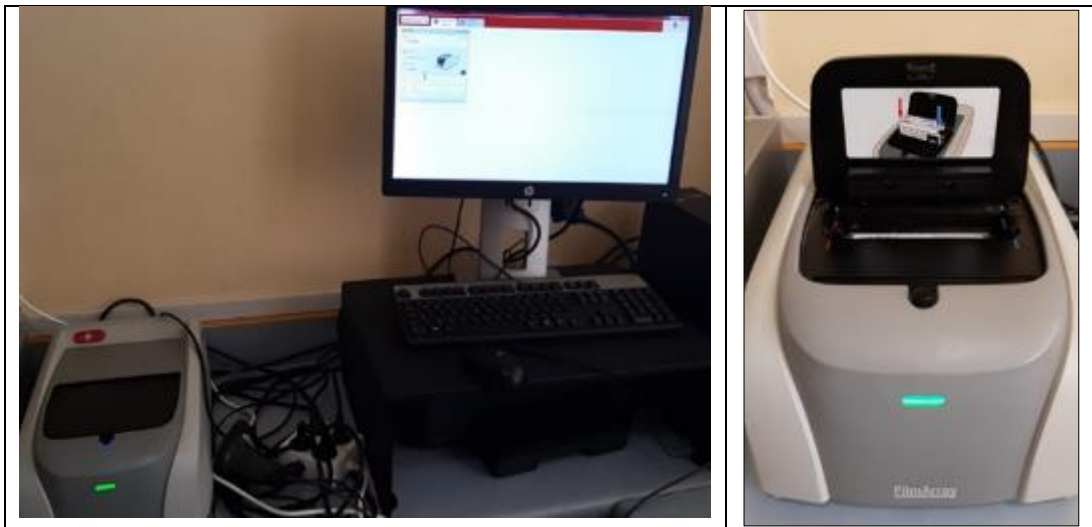
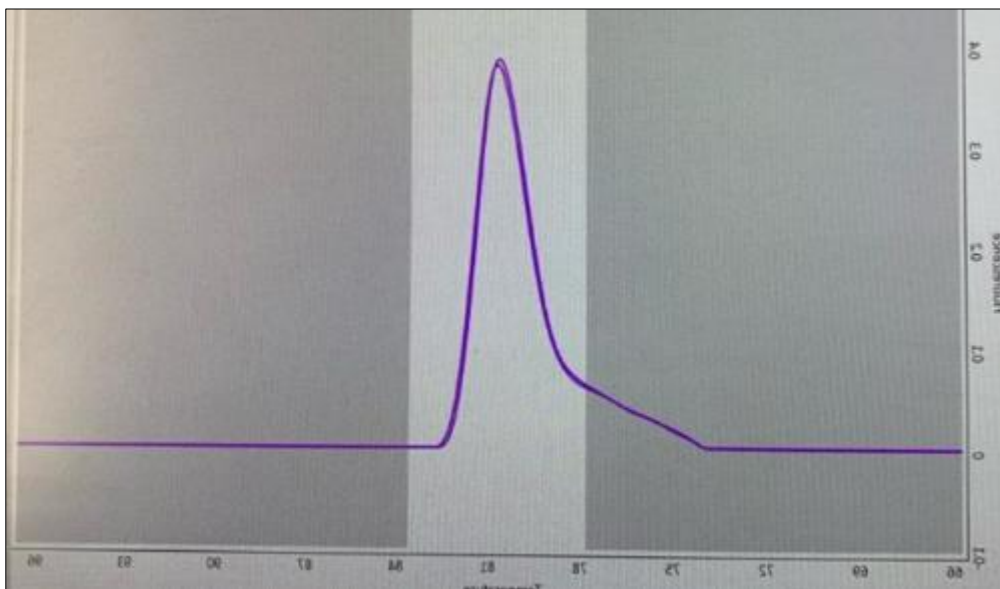
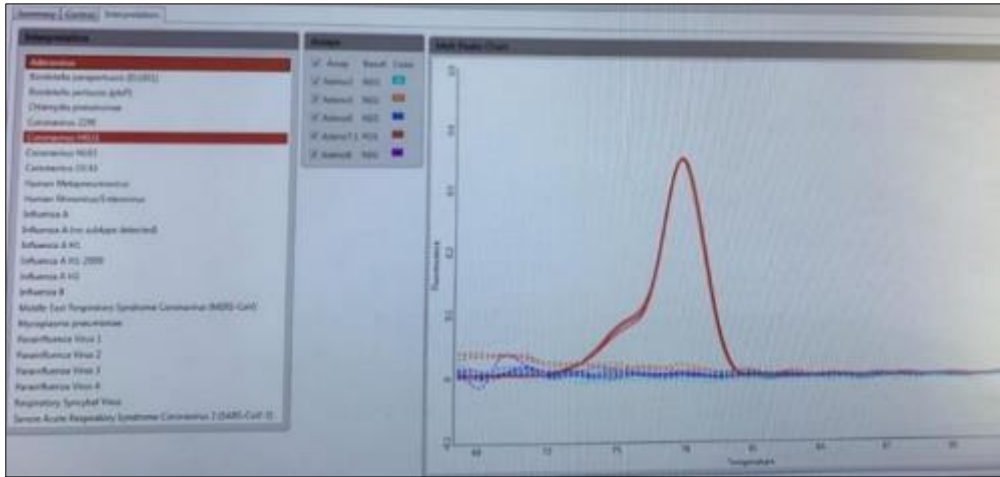


Figure 1 Images of the filmarray instrument with respiratory panel and positive curves

3 Results

In our study, 120 cases were evaluated, of which ninety patients were included, the remainder being excluded for lack of clinical data.

The mean age of the patients was 31 years, with extremes ranging from 1 month to 95 years, with a predominance of patients aged between 1 month and 20 years. Males were the most common, with a M/F sex ratio of 1.3.

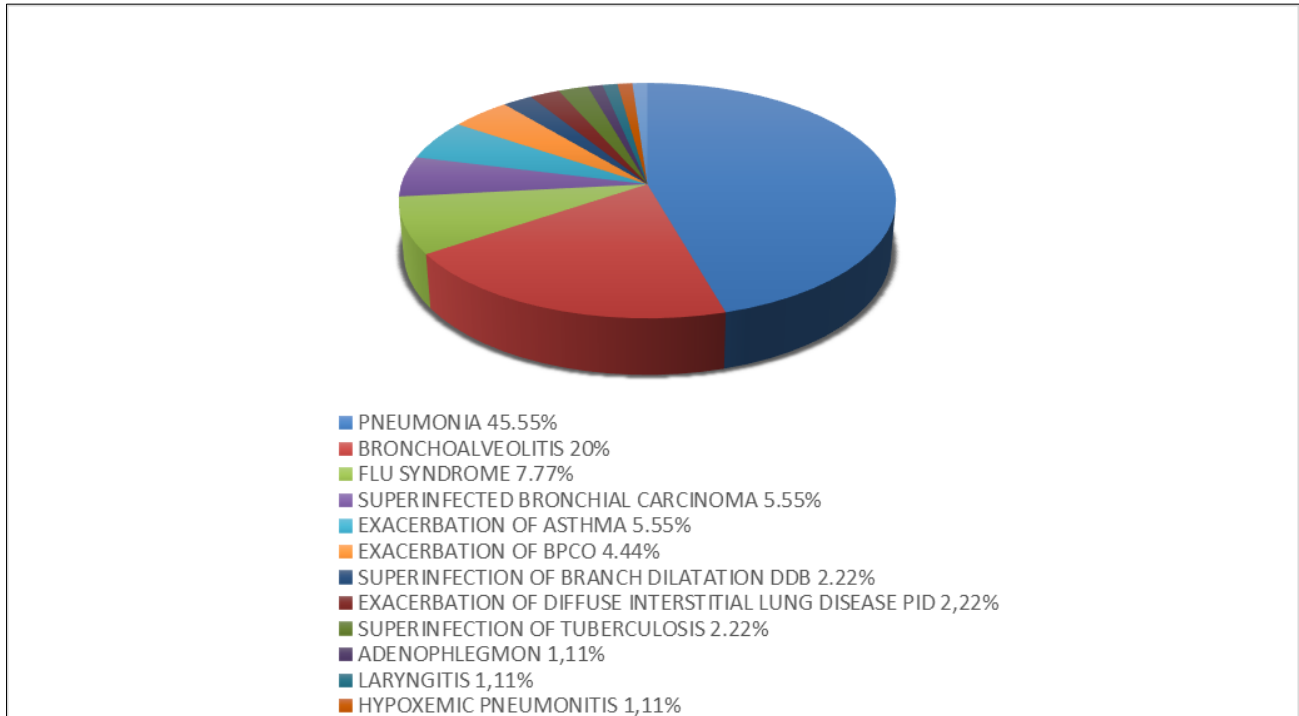


Figure 2 Breakdown of patient diagnoses

Pneumonia was the main diagnosis in 41 cases, followed by 18 cases of broncho alveolitis, 7 cases of influenza-like illness, 5 cases of superinfected bronchial carcinoma, 5 cases of asthma exacerbation, 4 cases of COPD exacerbation, 2 superinfections of DDB, exacerbation of PID, superinfection of tuberculosis, as well as one case of adenophlegmon, laryngitis, hypoxemic pneumonitis, and pneumocystis (Figure 2).

Respiratory PCR was negative in 26 cases (29%), positive for viral infection in 54 cases (60%), and positive for bacterial infection in 10 cases (11%).

Influenza A virus predominated in 19 cases, followed by Respiratory Syncytial Virus in 15 cases, adenovirus in 10 cases, SARS COV 2 in 8 cases, 4 cases of influenza B virus, 3 cases of HUMAN RHINOVIRUS/ ENTEROVIRUS, 3 cases of HKU1 coronavirus, 2 cases each of Para influenza virus 2 and 3 and one case of subtype 1, 2 cases of Human Metapneumovirus and one case of coronavirus 229E (Figure 3).

Acinetobacter baumannii complex was the most frequent multi-resistant bacterium in our series, with 7 cases, followed by *Staphylococcus aureus* with 4 cases, 3 cases of *Enterobacter cloacae* complex, 2 cases of *Pseudomonas aeruginosa*, *Escherichia coli*, *Klebsiella pneumoniae* and 1 case of *Proteus spp* (Figure 4).

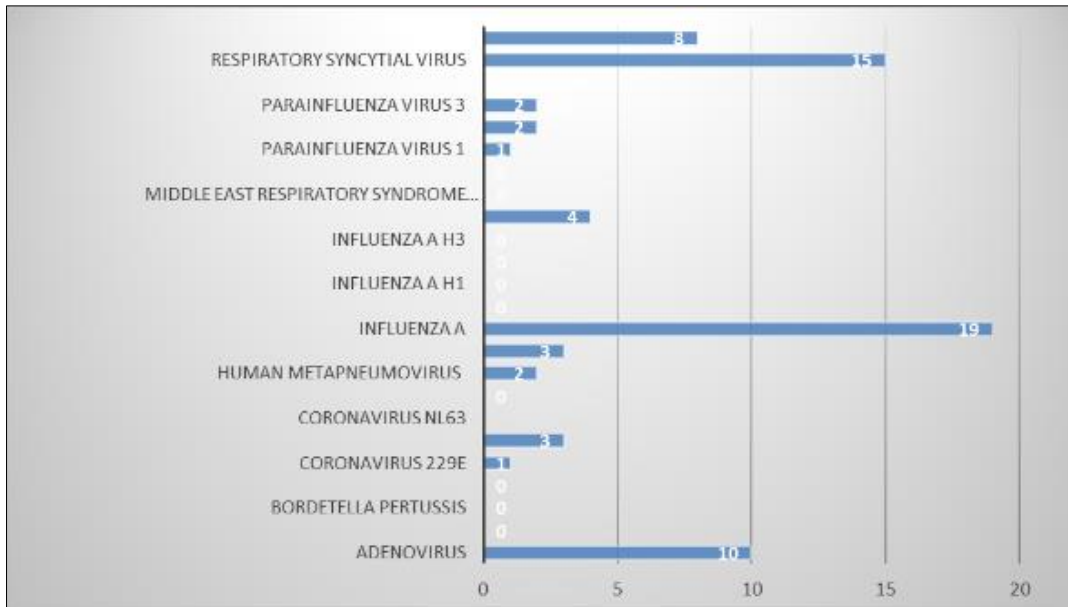


Figure 3 Distribution of RP 2.1 plus viral panel results

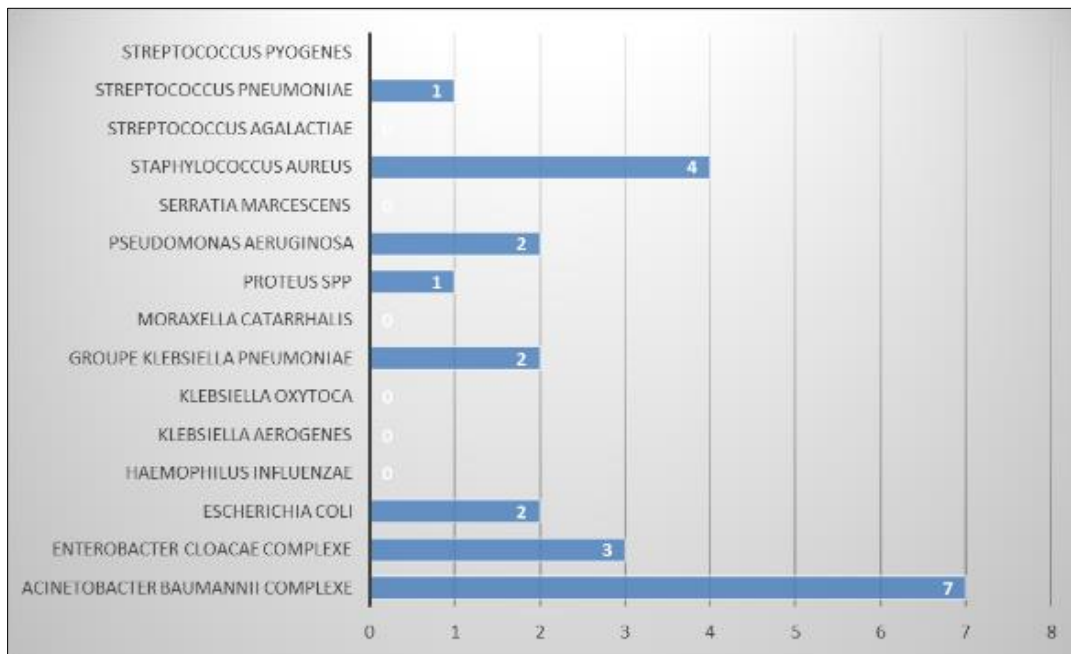


Figure 4 Distribution of RP 2.1 plus bacteria panel results

4 Discussion

The syndromic approach is a new method for diagnosing infectious diseases, which makes it possible to search simultaneously and in a single test for all the microorganisms most frequently responsible for an infection. We now know that mortality is directly correlated with delays in treatment and ineffective antibiotic therapy, and that overuse of antibiotics is the main epidemiological factor responsible for changing the microbiological profile through the emergence of resistance [3].

From the data in the literature, we can say that the microbial ecology is highly variable during lower respiratory infections, and the frequency of germs found varies in time and space, with a preponderance of approximately 70% to

90% of bacteria: Streptococcus Pneumoniae, Influenza A, Mycoplasma Pneumoniae and Hemophilus influenzae. Viruses responsible for 10% to 30% of acute community-acquired pneumonia come in second place [4].

The high prevalence of Parainfluenza A and RSV infection in our series points to a change in microbial ecology during this pandemic, a result close to that of the study, and thus in line with our hypothesis [5].

Previous studies have shown that a single multiplex PCR strategy is more cost-effective than an algorithm involving the use of successive techniques, which currently facilitates the management of COVID 19 infection [6].

This Film array pneumonia panel, which combines the detection of respiratory viruses and atypical bacteria with the semi-quantification of "classic" bacteria, a validated test on sputum and BAL, is currently being evaluated, particularly in patients hospitalised in intensive care for severe pneumonia. Initial results show that in 40% of cases it could lead to a reduction in the prescription of antibiotics initially prescribed [7]. In our series, most patients had a favourable outcome once treatment had been adapted.

New diagnostic technologies, the emergence of the SARS-CoV-2 coronavirus and the marketing of new antivirals will have an impact on recommendations for the management of these infections.

5 Conclusion

According to the literature, the germs responsible for respiratory infections vary and are bacterial, Streptococcus Pneumoniae, Influenza A, Mycoplasma Pneumoniae and Hemophilus Influenzae. According to our study, the COVID 19 pandemic could have a considerable and uneven impact on the ecology on which humanity depends. Current measures and methods of controlling this pandemic, from physical distancing to reinforced hygiene rules, could influence the diversity of the microbial ecology of lower respiratory infections. The wealth of literature on respiratory microbial ecology has not yet been developed following the COVID 19 pandemic, which is a subject of current research.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

Statement of ethical approval

The present research work does not contain any studies performed on animals/humans subjects by any of the authors.

Statement of informed consent

Informed consent was obtained from all individual participants included in the study.

References

- [1] Ramanan P, Bryson A.L, Binnicker M.J, et al: Syndromic Panel-Based Testing in Clinical Microbiology. Clin Microbiol Rev 2017; 31: e00024-17.
- [2] Multiple PCR panels (by TEM-PCRTM) for the detection of infectious diseases. Eurofins. Biomnis.
- [3] Virological diagnosis of lower respiratory infections S. Pillet M. Gueudin, J.C. Plantieret A. Revue des Maladies Respiratoires, 2021-01-01, Volume 38, Number 1, Pages 58-73.
- [4] Alimi Y., Lim W.S., Lansbury L., et. al: Systematic review of respiratory viral pathogens identified in adults with community-acquired pneumonia in Europe. J Clin Virol 2017; 95: pp. 26-35.
- [5] Contribution of multiplex PCR in describing the microbial ecology of respiratory infections during the COVID-19 pandemic M. Bougadoum, S. Ait Batahar, and L. Amro. Marrakech.
- [6] Mahony J.B., Blackhouse G., Babwah J., et al: Cost analysis of multiplex PCR testing for diagnosing respiratory virus infections. J Clin Microbiol 2009; 47: pp. 2812-2817.
- [7] 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: pp. 434.