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Original Article

Performance of a multiplex PCR pneumonia panel for the identification of respiratory pathogens and the main determinants of resistance from the lower respiratory tract specimens of adult patients in intensive care units

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Abstract *Background:* Timely diagnostic investigation to establish the microbial etiology of pneumonia is essential to ensure the administration of effective antibiotic therapy to individual patients.
Methods: We evaluated a multiplex PCR assay panel, the FilmArray® pneumonia panel (FilmArray PP, BioFire Diagnostics), for detection of 35 respiratory pathogens and resistance determinants and compared the performance of the standard-of-care test in intensive care unit patients with lower respiratory tract infections.
Results: Among the 59 endotracheal aspirates and bronchoalveolar lavage specimens obtained from 51 adult patients, FilmArray PP was effective in detecting respiratory bacterial pathogens with an overall positive percent agreement of 90% (95% confidence interval [CI], 73.5—97.9%) and negative percent agreement of 97.4% (95% CI, 96.0—98.4%). FilmArray PP semi-quantitative reporting demonstrated a concordance rate of 53.6% for the culture-positive specimens and 86.3% for the culture-negative specimens. FilmArray PP detected 16 viral

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targets, whereas the conventional viral isolation failed, except influenza A, which showed 100% concordance with PCR. Coinfections were detected in 42.3% of the specimens. Substantial discrepancies were observed in identifying antimicrobial resistance gene targets and in the susceptibility testing. However, FilmArray PP may still be useful at the early stage of pneumonia before culture and susceptibility test reports are available. Consequently, the results of FilmArray PP might alter the antibiotic prescription in 40.7% of the patients.
Conclusions: FilmArray PP offers a rapid and sensitive diagnostic method for lower respiratory tract infections. However, clinical correlation is advised to determine its significance in interpreting multiple pathogens and detection of genes involved in antimicrobial resistance.

Introduction

Pneumonia is a leading cause of hospitalization and death globally. According to the latest available data from the World Health Organization (WHO), lower respiratory tract infection is the fourth most common cause of mortality, causing 3.0 million deaths worldwide in 2016.[1] A delay in antibiotic administration can adversely affect the prognosis of pneumonia. Guidelines recommend early initiation of antibiotics in patients with pneumonia as several studies have suggested survival benefit when antibiotics are administered within 4 h of presentation.[2–4] Diagnostic investigations establishing the microbial etiology of pneumonia are essential to ensure the administration of effective antibiotics to the patients.[5] However, current therapy is typically initiated on an empirical basis, as even with the best diagnostic methods, a causative pathogen is often not detected in a significant proportion of pneumonia episodes. The Etiology of Pneumonia in the Community (EPIC) study reported a pathogen detection rate of 38% in patients who were hospitalized for pneumonia using traditional diagnostic techniques, including standard culture, antigen detection assay, and nucleic acid detection tests.[6] Such a low detection yield certainly calls for a more timely and sensitive diagnostic tool in pathogen identification. Nonetheless, differentiating the isolation of true pathogens and coincidental carriage strains in lower respiratory tract infections remains a challenge.[7] Over the last few years, molecular investigations have emerged as the diagnostic tool of choice for respiratory pathogens, particularly viruses, owing to its superior sensitivity in detecting organisms that are difficult to isolate, less viable, or present in only small numbers.[8] They are also less likely to be affected by prior antibiotic administration than those affected by standard culture methods.[9] In addition, molecular diagnostics may provide additional information regarding the presence of antibiotic resistance genes, which may promise better-targeted antimicrobial therapy and improve antibiotic stewardship.

The FilmArray® pneumonia panel (FilmArray PP, BioFire Diagnostics, Salt Lake City, UT, USA), a multiplex PCR assay panel, is an FDA-approved multiplex PCR assay that allows rapid and comprehensive detection of a wide range of clinically relevant targets and resistance markers from sputum (including endotracheal aspirate) and bronchoalveolar lavage (BAL) specimens. The target list includes 15 bacteria, 3 atypical bacteria, 8 viruses, and 7 antibiotic resistance genes. The panel also provides semi-quantitative results for 15 bacteria which may aid in distinguishing clinically relevant pathogens from colonizing bacteria and normal flora based on the estimates of relative nucleic acid abundance.[10] In this study, we first evaluated the clinical performance of FilmArray PP in 59 specimens from adults with lower respiratory tract infection and compared with the performance of the standard of care diagnostic investigation as requested by the primary care physicians.

Material and methods

Study subjects and clinical specimens

This study included 59 sputum (endotracheal aspirates) and BAL specimens collected between March 2019 and June 2019 from patients admitted to the medical intensive care units (MICU) at the National Taiwan University Hospital (NTUH) for respiratory failure. The hospital is a 2500-bed tertiary referral hospital located in northern Taiwan catering to a clinically diverse and complex patient population, including immunocompromised hosts undergoing solid organ or hematopoietic stem cell transplantation, and patients with cancer, cirrhosis, dialysis, or immunodeficiency. Most of the enrolled subjects were intubated; hence, the specimens were collected from endotracheal aspirates rather than expectorated sputum, which may be contaminated with the normal flora or colonizers from the upper respiratory tract. All the specimens were transported to the Clinical Microbiology Laboratory of the Hospital for analysis. The median time from specimen collection to loading into the FilmArray pouch was 1 h and 8 min (range, 12 min to 7 h and 30 min). A chart review was performed to determine the type and duration of antibiotic therapy in each subject. This study was approved by the Institutional Review Boards and Ethical Committees of the National Taiwan University Hospital (Taipei, Taiwan) [201903009RIND].

Microbiological cultures and molecular investigations

All sputum specimens were subjected to Gram staining and cultured according to the standard protocols to detect the

common respiratory pathogens. Conventional cultures were performed by inoculating a blood/eosin methylene blue agar and chocolate agar and incubating in an atmosphere enriched with 5% CO_2 at 35 °C. The culture plates were read at 18–24 h and held for 2 days before reporting as negative. The isolates were analyzed using standard biochemical methods and a Bruker Biotyper matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) system. Susceptibility testing was performed with the determination of the minimum inhibitory concentrations (MIC) using the Vitek2 platform (bioMérieux Inc., Durham, NC). Adenovirus, parainfluenza virus, respiratory syncytial virus, and *Chlamydia* were identified by direct immunofluorescence staining (Oxoid, Imagen, USA). The serum level of *Mycoplasma pneumoniae* IgM was determined using a rapid immunochromatographic test (Biocard™, AniBiotech, Finland), and serum *M. pneumoniae* IgG was assayed using an enzyme-linked immunosorbent assay (CHORUS kit, Diesse Diagnostica Senese, Italy). Urine specimens for *Legionella* and pneumococcal antigen detection were performed using the Alere BinaxNOW urinary antigen card (MA, USA). Influenza A and B were detected by fluorescent immunoassay (Sofia, Quidel, USA) using nasopharyngeal swab specimens.

FilmArray PP

The FilmArray PP is a syndrome-specific, cartridge-based, multiplex PCR that includes all steps of molecular diagnostics in an automated manner. An aliquot of each specimen was analyzed according to the manufacturer's instructions. The results were obtained in approximately 1 h. The pneumonia panel is compatible with all other FilmArray panel platforms. The principle and procedure of the assay have been described previously.[11] The panel included 15 bacteria (*Acinetobacter calcoaceticus baumannii complex, 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,* and *Streptococcus pyogenes*), 3 atypical bacteria (*Chlamydophilia pneumoniae, Legionella pneumophila,* and *M. pneumoniae*), 8 viruses (adenovirus, coronavirus, human metapneumovirus, human rhinovirus/enterovirus, influenza A, influenza B, parainfluenza virus, respiratory and syncytial virus), and 7 antimicrobial resistance genes (methicillin resistance *[mecA/C and MREJ*], carbapenemases [bla_{KPC}, bla_{NDM}, $bla_{OXA\text{-}48\text{-}like}$, bla_{VIM}, and bla_{IMP}], and ESBL [$bla_{CTX\text{-}M}$]). The results of the antimicrobial resistance genes (AMR) were reported qualitatively and conditionally if the potential microorganism of the gene is also detected in the sample. For example, *mecA/C* and *MREJ* were reported only in the presence of *S. aureus*. The detected resistance markers could not be linked to the detected microorganisms. The 15 bacteria were reported semi-quantitatively to the nearest whole log as genome copies/mL.

Data analyses

The FilmArray PP results were considered concordant when they were consistent with the results of conventional investigations. For the semi-quantitative analysis, a bin result of 10^4 or 10^5 was considered concordant with the standard-of-care result of "few", 10^5 or 10^6 was concordant with "moderate," and 10^6 or $>10^7$ was concordant with "many". Additionally, positive culture results that were not targets included in the FilmArray PP and results observed $<10^4$ copies/mL which were present in amounts too small for successful culture were both considered concordant. Otherwise, the results of FilmArray PP were discordant when they did not correspond with those of the standard-of-care investigation. The FilmArray PP result was considered true positive or true negative when it corresponded with the result of the standard-of-care investigation. A positive FilmArray PP result and a negative culture represented "false-positive," whereas a positive culture and negative FilmArray PP was is considered "false-negative." The positive percent agreement (PPA) was calculated as [true positive/(true positive + false negative)] × 100%, and negative percent agreement (NPA) was calculated as [true negative/(true negative + false positive)] × 100%. The performance measures of PPA and NPA only referred to bacterial analytes for which the gold standard of the bacterial culture was used as the reference method.

Results

Patient characteristics and specimen types

A total of 59 endotracheal aspirates and BAL specimens from 51 adult patients admitted to the MICUs were included in this study. For the seven patients who underwent bronchoscopy, both the specimens obtained from the endotracheal tube and bronchoscopy were sent for analysis. Among the patients, 43.9% were male and the median age was 65 years (range 30–97 years). The respiratory specimens comprised of 40 sputum (endotracheal aspirate included) specimens, 13 BAL, and 6 bronchial washing specimens.

Findings of FilmArray PP

The FilmArray PP detected at least one pathogen in 33 of the 59 specimens that were tested, yielding a positivity rate of 55.9%. Table 1 summarizes the total pathogens and resistance genes detected in the study. The panel identified a potential pathogen in 47.4% of BAL and bronchial wash specimens and 60% of the sputum specimens. The most prevalent pathogens detected were *K. pneumoniae* (21.3%), *P. aeruginosa* (14.9%), *A. calcoaceticus baumannii* complex (12.8%), and *S. aureus* (12.8%). Co-infections (including virus) were detected in 14 (42.4%) of the positive specimens and the greatest number of pathogens detected in a single specimen was eight (*Acinetobacter baumannii* complex, *E. aerogenes, E. cloacae* complex, *E. coli, K. pneumoniae, P. aeruginosa, S. marcescens,* and *S. aureus*). Multiple detections per single specimen were higher in sputum (92.9%) specimens than in BAL and bronchial wash (7.1%) specimens (Table 2). Viruses were detected in 16 (27.1%) specimens; influenza A was the most commonly detected virus (8.5%), followed by adenovirus and human metapneumovirus (5.1%) (Table 1). Viruses and bacteria were observed together in 10.2% (n = 6) of the specimens, and six viruses (adenovirus, coronavirus, human

Table 1 Total number of FilmArray pneumonia panel detections (n = 59).

Pathogen	Total no. (%)
Bacteria	
Any bacteria	24 (40.7)
Acinetobacter calcoaceticus baumannii complex	6 (10.2)
Enterobacter cloacae complex	3 (5.1)
Escherichia coli	4 (6.8)
Haemophilus influenzae	3 (5.1)
Klebsiella aerogenes (E. aerogenes)	1 (1.7)
Klebsiella oxytoca	1 (1.7)
Klebsiella pneumoniae	10 (16.9)
Legionella pneumophila	2 (3.4)
Pseudomonas aeruginosa	7 (11.9)
Serratia marcescens	3 (5.1)
Staphylococcus aureus	5 (8.5)
Streptococcus agalactiae	2 (3.4)
Streptococcus pneumoniae	1 (1.7)
Viruses	
Any virus	16 (27.1)
Adenovirus	3 (5.1)
Coronavirus	1 (1.7)
Human metapneumovirus	3 (5.1)
Human rhinovirus/enterovirus	2 (3.4)
Influenza A virus	5 (8.5)
Parainfluenza virus	2 (3.4)
Resistance genes	
Carbapenemases	3 (5.1)
bla_{IMP}	1 (1.7)
bla_{NDM}	1 (1.7)
bla_{VIM}	1 (1.7)
ESBL	
bla_{CTX-M}	5 (8.5)

rhinovirus/enterovirus, influenza A, parainfluenza virus) were detected from the bronchial aspirate or BAL specimens. Multiple viruses were not observed in any of the specimens. *M. catarrhalis*, *Proteus* spp., *S. pyrogenes*, *C. pneumoniae*, *M. pneumoniae*, influenza B, and respiratory syncytial virus were not detected in this study. Among the respiratory specimens tested, four bla_{CTX-M} extended-spectrum β-lactamases (ESBL) and three carbapenemases genes (bla_{IMP}, bla_{NDM}, and bla_{VIM}) were detected.

Evaluation of the performance of filmarray PP

The performance data for each FilmArray PP target are summarized in Tables 3 and 4. The PPA and NPA with a 95% confidence interval (CI) of the FilmArray PP with reference investigations were 90% (73.5—97.9) and 97.4% (96.0—98.4), respectively. Overall, there were 37 concordant specimens and 10 discordant specimens, yielding an overall agreement of 79%. FilmArray PP showed a concordance rate of 100% for five targets as follows: *E. cloacae complex*, *E. coli*, *H. influenzae*, *S. marcescens*, and *S. pneumoniae*. FilmArray PP detected *L. pneumophila* in two specimens but serological tests failed to identify both of them. One sputum sample yielded *K. oxytoca*, which was not reported on the FilmArray PP. Five analytes demonstrating a concordance rate of <90% as follows *A. calcoaceticus baumannii complex* (83%), *K. pneumoniae group* (80%), *P. aeruginosa* (71%), *S. aureus* (80%), and *S. agalactiae* (50%). There was no *M. catarrhalis*, *Proteus* spp., *S. pyogenes*, *C. pneumoniae*, and *M. pneumoniae* detected in this study, and therefore, no PPA and NPA could be calculated. Notably, FilmArray PP was able to detect bacteria in 7 out of 29 (24.1%) culture-negative specimens. Specimens from 18 patients (30.5%) yielded bacteria that were not included in the FilmArray PP (*Burkholderia cepacia* complex, *Citrobacter freundii*, *Chryseobacterium indologenes*, *Enterococcus faecium*, *Morganella morganii*, *Raoultella ornithinolytica*, *Ralstonia manitolilytica*, *Sphingomonas paucimobilis*, and *Stenotrophomonas maltophilia*). The FilmArray PP semi-quantitative report demonstrated a concordance rate of 53.6% for the culture-positive specimens and 86.3% for the culture-negative ones (Table 5). Overestimation of quantification was observed which could be attributed to the detection of dead organisms by the FilmArray PP.

FilmArray PP detected significantly more viruses (adenovirus, coronavirus, human metapneumovirus, human rhinovirus/enterovirus, and parainfluenza virus) than the standard diagnostic method. Not all viruses were reported by viral culture or identification; however, influenza A demonstrated a 100% positive agreement with the nucleic acid test (including two BAL specimens). Of the four bla_{CTX-M} detected by FilmArray PP, only one case could be verified by the MIC method. As listed in Table 6, the corresponding pathogens were not detected by the standard culture, and thus, no further sensitivity test was performed. The three carbapenemases observed except for bla_{VIM} were consistent with the MIC method and conferred penicillin, cephalosporin, and carbapenem resistance. Two carbapenem-resistant *P. aeruginosa* and one *A. baumannii* were detected by culture, but no antimicrobial resistance gene was identified by FilmArray PP.

Impact on antimicrobial prescribing

The FilmArray PP results might lead to a de-escalation of initial empirical antibiotics in 16 (27.1%) patients,

Table 2 Number of pathogens per single specimen, data by specimen type.

Detections	Negative	Positive	1 analyte	2 analytes	3 analytes	4 analytes	5 analytes	6 analytes	7 analytes	8 analytes
BAL	7	6	6	0	0	0	0	0	0	0
Bronchial wash	3	3	2	1	0	0	0	0	0	0
Sputum	16	24	11	7	4	0	0	1	0	1
Total	26 (44.1%)	33 (55.9%)	19 (57.6%)	8 (24.2%)	4 (12.1%)	0 —	0 —	1 (3.0%)	0 —	1 (3.0%)

Table 3 Concordance rate of each bacterial analyte detected in the study.

Organisms	Concordant	Discordant	Concordance rate (%)
Acinetobacter calcoaceticus baumannii complex	5	1	83
Enterobacter cloacae complex	3	0	100
Escherichia coli	4	0	100
Haemophilus influenzae	2	1	67
Klebsiella aerogenes (E. aerogenes)	0	1	0
Klebsiella oxytoca	0	1	0
Klebsiella pneumoniae	8	2	80
Legionella pneumophila	0	2	0
Pseudomonas aeruginosa	5	2	71
Serratia marcescens	3	0	100
Staphylococcus aureus	4	1	80
Streptococcus agalactiae	1	1	50
Streptococcus pneumoniae	1	0	100
All	36	12	75

Table 4 Performance summary for bacterial analytes detected by FilmArray pneumonia panel and routine culture.

Organisms	FilmArray [b] Culture [b]	FilmArray [b] Culture -	FilmArray -Culture [b]	FilmArray -Culture -	PPA[a] % (95% CI)	NPA[a] % (95% CI)
Acinetobacter calcoaceticus baumannii complex	4	2	0	53	100 (39.8—100)	96.4 (87.5—99.6)
Enterobacter cloacae complex	2	1	0	56	100 (15.8—100)	98.3 (90.6—100)
Escherichia coli	3	1	0	55	100 (29.2—100)	98.2 (90.5—100)
Haemophilus influenzae	0	3	0	56	—	94.9 (85.9—98.9)
Klebsiella aerogenes (E. aerogenes)	0	1	0	58	—	98.3 (90.9—100)
Klebsiella oxytoca	0	0	1	58	—	100 (93.8—100)
Klebsiella pneumoniae group	6	3	1	49	85.7 (42.1—99.6)	94.2 (84.1—98.8)
Legionella pneumophila[b]	0	2	0	57	—	96.6 (88.3—99.6)
Pseudomonas aeruginosa	5	1	1	52	83.3 (35.9—99.6)	98.1 (89.9—100)
Serratia marcescens	2	1	0	56	100 (15.8—100)	98.3 (90.6—100)
Staphylococcus aureus	4	2	0	53	100 (39.8—100)	96.4 (87.5—99.6)
Streptococcus agalactiae	1	1	0	57	100 (2.5—100)	98.3 (90.8—100)
Streptococcus pneumoniae	0	1	0	58	—	98.3 (90.9—100)
All	27	19	3	718	**90 (73.5—97.9)**	**97.4 (96.0—98.4)**

[a] PPA, positive percent agreement; NPA, negative percent agreement.
[b] PPA and NPA were calculated with respect to standard culture for bacteria analytes and serology testing for atypical pathogens.
There was no *Moraxella catarrhalis*, *Proteus* spp., *Streptococcus pyogenes*, *Chlamydophilia pneumonia*, and *Mycoplasma pneumoniae* detected in this study.

escalation or addition of another effective antibiotic in 9 (13.6%) patients, and no change in 33 (55.9%) patients.

Discussion

The FilmArray PP combines nested multiplex PCR and real-time PCR amplification data by performing nucleic acid extraction, amplification, detection, and data analysis in a disposable pouch system to provide a semi-quantitative report.[12] This multiplex molecular assay was recently FDA-approved for the identification of 33 respiratory targets within 1 h to aid in the diagnosis of lower respiratory tract infections.

In this prospective study, the performance characteristics of FilmArray PP were estimated by measuring its agreement with the standard-of-care diagnostic investigation. The overall performance of FilmArray PP was comparable to other multiplex respiratory platforms with an overall agreement of >80% for all the available targets

Table 5 FilmArray pneumonia panel semi-quantitative results (bin values) as compared to the standard-of-care results.

Standard-of-care level		Culture			
		Not reported	Few	Moderate	Many
FilmArray bin (copies/ml)	Not detected	35	2	1	0
	10^4	9	3	3	0
	10^5	3	3	4	0
	10^6	4	6	3	1
	10^7	0	0	1	1
% concordant		44/51	6/14	7/12	2/2
		86.3%	42.9%	58.3%	100%
			53.60%		

Grey shades indicate the expected bin results based on the analyte concentration.

tested.[13,14] Most of the bacterial targets detected were Gram-negative pathogens (GNB) and viruses accounting for 27% of all cases. According to the local epidemiology data, the FilmArray PP may reach approximately 70—90% coverage for the most prevalent agents responsible for moderate to severe community-acquired pneumonia among adults in Taiwan, and 70—80% coverage for healthcare-associated pneumonia.[15—17] In line with our results, a systematic review of the etiologic agents of community-acquired pneumonia in Asia revealed a high prevalence of GNB and *S. aureus* as causative pathogens as compared to the commonly observed *S. pneumoniae* and *H. influenzae* in the western countries.[18,19] The incidence of respiratory tract infection caused by *S. pneumoniae* and *H. influenzae* has also reduced following implementation of vaccination programs among the elderly and children.[20,21] Additionally, discrepancies could have been caused by the fastidious nature of *H. influenzae*, which is difficult to culture.[22] In our study, three specimens of *H. influenzae* were observed by FilmArray PP but were not recovered by culture. Two of these specimens reported 10^4 copies/mL of bacteria which might be too low to yield a positive culture.

Both serology and culture methods failed to identify the two *L. pneumophila* found by FilmArray PP. The diagnosis of *Legionella* infection has been limited by difficulties in its culture and the non-specific nature of serological investigations, which only identifies *L. pneumophila* serogroup 1 but not the other species and serogroups.[23,24] Most of the discordant results (FilmArray PP positive but culture-negative) in our study were probably caused by antibiotic use prior to sampling. As the nucleic acid of an organism may persist *in vivo* independent of the organism's viability, the detection of a target does not indicate that the corresponding organisms are the causative agents for the particular clinical symptoms. Likewise, the performance of the FilmArray PP has not been established for monitoring the treatment of infection. Another challenge posed by molecular diagnostics for respiratory pathogens is the difficulty in distinguishing colonization from infection. Though the presence of non-colonizing organisms may suggest disease causation, certain respiratory pathogens such as *S. pneumoniae*, rhinovirus, or adenovirus may colonize in the upper airway of an asymptomatic individual, and thus, leading to a diagnostic dilemma.[5] Quantification of microbial load by molecular methods may provide some clues as isolates with greater quantities are more likely to be clinically significant.[9] Defining the true pathogen responsible for the disease is further complicated by the detection of co-infections. In this study, coinfections (both viral-bacterial and multiple bacterial pathogens) were identified in 42.3% of the specimens. Nevertheless, there is growing evidence regarding the incidence and pathogenesis of polymicrobial pneumonia[25—27] and the recent major revelation of the lung microbiome has challenged our traditional paradigm that lungs are sterile and that pneumonia is caused by a single invasive pathogen.[28,29] Future research on pneumonia thus needs to address the conundrum of polymicrobial respiratory disease and its impact on the pathogenesis of pneumonia.[29]

There were substantial discrepancies in the detection of antimicrobial resistance genes in our study. As there are multiple genetic variants and mechanisms of resistance, the detection of no resistance gene does not imply susceptibility to the associated antibiotic. Similarly, the presence of a specific genetic marker cannot be linked to the pathogen detected. Complete culture and susceptibility testing should, thus, be performed for each potential following the recommended guidelines.[30]

Several limitations to this study could be addressed of which the most notable one is the lack of a "gold-standard" reference method to clarify the discrepant results between FilmArray PP and conventional investigations. Multiplex PCR is more likely to have better sensitivity than traditional culture and serological investigation, and thus, leading to difficulties in interpreting the clinical significance of false-positive results.[8] Therefore it is inappropriate to consider a positive FilmArray PP result as false-positive when compared to a less-sensitive test.[5] The unavailability of an established reference hinders the true estimate of accuracy. Next, the patients enrolled in the study were highly heterogeneous. Patients with various causes of respiratory failure, including community-acquired pneumonia, healthcare-associated pneumonia, ventilator-associated pneumonia, or non-infectious lung disease, who required intubation for ventilatory support, were included in the study. While the respiratory specimens obtained from intubated patients are more representative of the site of infection and are less likely to be contaminated by upper airway flora, the pathogens responsible for each clinical context were different and may not be parts of the targets in the

Table 6 List of resistance genes detected by FilmArray pneumonia panel and conventional MIC methods. In the last three cases, the resistance markers were not detected with FilmArray for the corresponding phenotypes.

Age/gender	Bacterial culture	FilmArray results		Susceptibility testing results						
		Bacteria (copies/ml)	Resistance genes	FOX	CAZ	CTX	FEP	ETP	IPM	MEM
99/M	Yeast (1+)	*K. pneumoniae* (10^4)	bla_{CTX-M}							
87/F	*E. faecium* (1+)	*A. baumannii* complex (10^4) *K. pneumoniae* (10^4)	bla_{CTX-M}							
53/M	*K. pneumoniae* (few) *S. aureus* (2+)	*K. pneumoniae* (10^6) *S. aureus* (10^7)	bla_{CTX-M}	S (0.5)	S (≤1)	S (≤1)	S (≤1)	S (≤0.5)	S (0.5)	S (≤0.25)
85/F	*K. pneumoniae* (1+) *E. cloacae* (few) *P. aeruginosa* *E.coli* *C. freundii*	*A. baumannii* complex (10^5) *E. aerogenes* (≥10^5) *E. cloacae* complex (10^6) *E. coli* (10^6) *K. pneumoniae* (10^6) *P. aeruginosa* (10^6) *S. marcescens* (10^4) *S. aureus* (10^6)	bla_{CTX-M}		R (16) S (≤1) S (4) R (16) S (≤1)	I (2) S (≤1) R (32) R (4) S (≤1)	S (≤1) S (≤0.5) S (4) S (≤1) S (≤1)	S (≤0.5) S (≤0.25) S (≤0.5) S (≤0.5)	S (≤0.25) S (≤0.25) S (2) S (≤0.25) S (0.5)	S (≤0.25) S (≤2) S (≤0.25) S (≤0.25) S (≤0.25)
64/M	*E. coli* (1+) *S. maltophilia* (2+)	*E. coli* (10^5)	bla_{VIM}		S (≤1)	S (≤1)	S (≤1)	S (≤0.5)	S (≤02.5)	S (≤02.5)
84/F	*A. pittii* (few) *Ralstonia mannitolilytica* (1+)	*A. baumannii* complex (10^4)	bla_{IPM}, bla_{NDM}		R (≥64)		R (≥64)		R (≥16)	R (≥16)
88/F	*P. aeruginosa* (2+) *K. oxytoca* (1+) *S. maltophilia* (1+)	*P. aeruginosa* (10^6)			S (8) R (16)	R (≥64) R (4)	S (8) S (≤1)	 S (≤0.5)	R* (2*) S (≤0.25)	I (4) R (4*)
53/M	*A. baumannii* (few) *S. paucimobilis* (few)	*A. baumannii* complex (10^5) *P. aeruginosa* (10^6)			R (64) I (16)	R (64) S (2)	R (64) S (8)		R (≥16) S (≤0.25)	R (≥16) S (≤02.5)
82/M	*P. aeruginosa* (few)	None			R (≥64)	R (≥64)	R (≥64)		R (8)	I (4)

FOX, cefoxitin; CAZ, ceftazidime; CTX, cefotaxime; FEP, cefepime; ETP, ertapenem; IPM, imipenem; MEM, meropenem.

panel. For instance, nosocomial pathogens, such as *S. maltophilia*, *B. cepacia* complex, and *Elizabethkingia meningoseptica*,[31] might outgrow the common causative bacteria as a result of antibiotic selection, thereby leading to under-detection as they were not included in the panel. Some pathogens, such as *Pneumocystis jiroveci* and non-tuberculous mycobacteria that were also not included in the panel, commonly cause respiratory infections in immunocompromised hosts in Taiwan.[32–35] However, this enables us to understand the application of the FilmArray PP across different patient populations.

In summary, the FilmArray PP may provide early information on the causative pathogens and their determinants of resistance, allowing a pathogen-directed antibiotic therapy with 70—90% coverage for the most prevalent bacteria causing moderate to severe community-acquired pneumonia among the adults in Taiwan. Although FilmArray PP may not replace conventional culture and antimicrobial susceptibility testing, especially for bacterial targets, it is still an efficient adjunct to guide clinical decisions and antibiotic treatment in the early stages of pneumonia.

Conflicts of interest

None declared.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jmii.2019.10.009.