J Infect Chemother xxx (2018) 1–5

Contents lists available at ScienceDirect

Journal of Infection and Chemotherapy

journal homepage: http://www.elsevier.com/locate/jic

Original Article

Evaluation of FilmArray respiratory panel multiplex polymerase chain reaction assay for detection of pathogens in adult outpatients with acute respiratory tract infection

Norihito Kaku [a, *], Kohji Hashiguchi [b], Yuki Iwanaga [a], Norihiko Akamatsu [a], Junichi Matsuda [a], Kosuke Kosai [a], Naoki Uno [a], Yoshitomo Morinaga [a], Takeshi Kitazaki [b], Hiroo Hasegawa [a], Taiga Miyazaki [c], Masaaki Fukuda [b], Koichi Izumikawa [d], Hiroshi Mukae [c], Katsunori Yanagihara [a]

[a] Department of Laboratory Medicine, Nagasaki University Graduate School of Biomedical Sciences, Nagasaki, Japan
[b] Department of Respiratory Medicine, Japanese Red Cross Nagasaki Genbaku Hospital, Nagasaki, Japan
[c] Department of Respiratory Medicine, Nagasaki University Graduate School of Biomedical Sciences, Nagasaki, Japan
[d] Department of Infectious Diseases, Nagasaki University Graduate School of Biomedical Sciences, Nagasaki, Japan

ARTICLE INFO

Article history:
Received 9 December 2017
Received in revised form
1 May 2018
Accepted 16 May 2018
Available online xxx

Keywords:
Respiratory infection
Viral infection
Influenza
Molecular biology

ABSTRACT

Although viruses are the major pathogen that causes upper respiratory tract infection (URTI) and acute bronchitis, antibiotics have been prescribed. This was a prospective observational study in influenza epidemics that enrolled adult outpatients who visited a hospital with respiratory tract infection symptoms. In this study, we evaluated the usefulness of FilmArray respiratory panel (RP). Fifty patients were enrolled. FilmArray RP detected the pathogens in 28 patients. The common pathogens were *influenza virus* (n = 14), *respiratory syncytial virus* (n = 6), and *human rhinovirus* (n = 6). Of the 14 patients with *influenza virus*, 6 were negative for the antigen test. The physicians diagnosed and treated the patients without the result of FilmArray in this study. Of the patients with positive FilmArray RP, 9 were treated with antibiotics; however, bacteria were detected in only 3 patients. By implementing FilmArray RP, URTI and acute bronchitis would be precisely diagnosed, and inappropriate use of antibiotics can be reduced.

1. Introduction

Acute respiratory tract infection (ARTI) is one of the major infectious diseases that may occur at any age and accounts for 3.5 million deaths worldwide [1]. ARTIs are classified as acute upper respiratory tract infection (URTI), acute bronchitis, or pneumonia. Diagnosis of ARTI except pneumonia is largely based on clinical signs and symptoms, because viruses, the most commonly causative pathogens of URTI and acute bronchitis, are difficult to detect. However, antibiotics have been prescribed in many patients with URTI or acute bronchitis [2–4]. Inappropriate prescription of antibiotics promotes antibiotic resistance. Therefore, these viruses should be detected.

These viruses could be detected using rapid antigen determination tests; however, their sensitivity was relatively low, e.g., the pooled sensitivity of influenza antigen test in adults and children with influenza-like illness was 62.3% [5]. To improve the sensitivity in detecting the viruses, the nucleic acid amplification test (NAAT) can be used. The NAAT has been developed for various viruses [6] and could detect multiple targets [7]. Despite these advantages, the use of NAAT has been infrequent because of its complicated procedures and difficulty in performing at community hospitals.

In the past few years, several fully automated platforms for NAAT were developed. These platforms can be performed simply, provide rapid results, and are used as an assay for multiple organisms from a single sample. In this study, one of the fully automated platforms, the FilmArray® Respiratory panel (RP), was evaluated. FilmArray® RP targets 20 pathogens, including 17 viruses and subtypes and 3 bacteria, and is performed in approximately 1 h turn-around-time in adult outpatients with ARTI.

* Corresponding author. Department of Laboratory Medicine, Nagasaki University Graduate School of Biomedical Sciences, 1-7-1 Sakamoto, Nagasaki 852-8501, Japan.
E-mail address: kaku-ngs@umin.ac.jp (N. Kaku).

https://doi.org/10.1016/j.jiac.2018.05.006
1341-321X/

2. Material and methods

2.1. Study design

A prospective observational study was conducted between January 15 and April 5, 2016, in Nagasaki University Hospital and Nagasaki Genbaku Hospital. We enrolled adult outpatients who visited the Department of Respiratory Medicine and the Japanese Red Cross Nagasaki Genbaku Hospital with respiratory tract infection symptoms such as cough, sputum, sore throat, nasal mucus, headache, dyspnea, or hypoxemia. Based on the physician's discretion, chest X-ray and microorganism tests, such as gram stain, culture, and influenza antigen test, were performed at the Japanese Red Cross Nagasaki Genbaku Hospital. Informed consent and nasopharyngeal swabs for FilmArray RP were obtained from all patients. The FilmArray RP analysis was performed at Nagasaki University Hospital; however, the results were not reported to the physicians.

2.2. Diagnostic criteria

The physicians determined the clinical diagnosis without the FilmArray RP results. Patients with abnormal shadow in the chest X-ray were diagnosed with pneumonia. The classification of URTI and acute bronchitis were determined based on the clinical history and findings of the two physicians, who are certified board members of the Japanese Respiratory Society.

2.3. Influenza antigen test

Influenza antigen test was performed at the Japanese Red Cross Nagasaki Genbaku Hospital. In most cases, BD Veritor System™ for rapid detection of Flu A + B (Becton, Dickinson and Company, New Jersey, USA) was performed as recommended by the manufacturer. ImmunoAce Flu (TAUNS Laboratories, Inc., Sizuoka, Japan) was performed as recommended by the manufacturer for some patients who visited the hospital after consultation hours.

2.4. FilmArray RP

FilmArray RP was supplied by the SYSMEX bioMérieux Co., Ltd. (Tokyo, Japan). It includes assays that detect *Adenovirus*; *Coronavirus* (229E, HKU1, OC43 and NL63); *Human metapneumovirus*; *Human rhinovirus*; *enterovirus*; Influenza A with specific detection of subtypes H1, H1-2009, and H3; Influenza B, Parainfluenza types 1 to 4; *Respiratory syncytial virus*; *Chlamydophila pneumoniae*; *Mycoplasma pneumoniae*; and *Bordetella pertussis*. Testing was performed at Nagasaki University Hospital as recommended by the manufacturer.

2.5. Quantitative reverse transcriptase polymerase transcription assay (qRT-PCR)

In two samples, FilmArray RP detected only one gene (FluA-pan2), and their results were “equivocal.” In these samples, further genetic analysis using the qRT-PCR was performed as previously reported [8], because there was a possibility of a false positive or negative for *Influenza virus* A. A one-step qRT-PCR was performed using LightCycler 480 RNA Master Hydroas [8]. RT-PCR was performed at 63 °C for 3 min and 95 °C for 30 s, followed by 45 cycles at 95 °C for 10 s and 58 °C for 30 s. Standard curves were drawn from serial dilutions of viral RNA standards.

2.6. Ethics

This study was approved by the ethics committee of Nagasaki University Hospital (approval number, 15122108) and the Japanese Red Cross Nagasaki Genbaku Hospital (approval number, 413). This study was registered at UMIN-CTR (reference number: UMIN000026464).

2.7. Statistical analysis

A statistical software package (StatMate V; ATMS Co., Ltd., Tokyo, Japan) was used for all the statistical comparisons, which were all two-tailed unpaired and tests of significance. The statistical significant α-level was set as ≤ 0.05. The chi-square or Fisher's exact test was used to compare categorical variables.

3. Results

3.1. Patient characteristics

During the study period, a total of 50 patients (22 men and 28 women) with respiratory tract symptoms were evaluated. Patient characteristics were shown in Table 1. The mean, maximum, and minimum age of the patients were 63.1 ± 20.0, 89, and 24 years, respectively. Among the study patients, 29 (58%) had underlying diseases: bronchial asthma (10, 20%); COPD (5, 10%); hypertension (4, 8%); Bronchiectasis (3, 6%); Diabetes mellitus (3, 6%); other respiratory diseases (4, 8%); and other diseases (3, 6%). Common symptoms were fever (74%), cough (74%), sputum (48%), and nasal mucus (46%). In 23 patients (46%), abnormal respiratory sounds were auscultated. The most common microbiology test in patients was influenza antigen test (41, 82%). The positive rate of the influenza antigen test was 22.0% (n = 9). Sputum culture was conducted in 18 patients; 12 of them had positive results. The most common bacteria isolated was *Haemophilus influenzae*, which was detected in seven patients.

Table 1
Patient characteristics.

Age	63.1 ± 20.0	
Sex (male/female)	22/28	
Clinical symptoms	50	(100%)
Fever	37	(74%)
Cough	37	(74%)
Sputum	24	(48%)
Nasal mucus	23	(46%)
Sore throat	16	(32%)
Dyspnea	15	(30%)
Headache	12	(24%)
Hypoxemia	10	(20%)
Abnormal respiratory sounds	23	(46%)
Coarse crackles	14	
Wheezes	8	
Decreased breath sounds	2	
Chest X-ray	26	(52%)
Abnormal shadow	21	
Microbiology test		
Influenza antigen test	41	(82%)
positive	9	
Pneumococcal urinary antigen test	25	(50%)
positive	2	
Legionella urinary antigen test	24	(48%)
positive	0	
Sputum culture	18	(36%)
Positive	12	
Haemophilus influenzae	7	
Staphylococcus aureus	2	
Escherichia coli	2	
Others	5	

Please cite this article in press as: Kaku N, et al., Evaluation of FilmArray respiratory panel multiplex polymerase chain reaction assay for detection of pathogens in adult outpatients with acute respiratory tract infection, J Infect Chemother (2018), https://doi.org/10.1016/j.jiac.2018.05.006