

# Evaluation of the Luminex Diagnostics® Respiratory Viral Panel for the Detection of 16 Respiratory Viruses and Subtypes in Children

J.G. Newland, R. Selvarangan, E.A. Thorell, C. J. Harrison  
Children's Mercy Hospitals and Clinics, Kansas City, MO.



## ABSTRACT (REVISED)

### Background:

Molecular amplification techniques have been shown to offer higher sensitivities than conventional viral diagnostics. The respiratory viral panel (RVP) from Luminex Diagnostics is a multiplex nucleic acid amplification test (NAT) that can detect 16 different viruses and subtypes from a single respiratory specimen. The RVP detects respiratory syncytial virus (RSV), influenza A (Flu A), influenza B (Flu B), adenovirus (Ad), parainfluenza 1, 2, 3, 4 (PIV 1,2,3,4), enterovirus (EV)/rhinovirus (RhV), coronavirus (CoV), and human metapneumovirus (HMPV).

### Methods:

A total of 140 frozen nasal aspirates from children were tested by RVP in two stages to evaluate its performance. (1) **Known Specimens:** To detect 44 viruses previously identified by culture techniques or rapid antigen testing. (2) **Unknown Specimens:** To detect respiratory viruses from 96 rapid antigen negative specimens that were simultaneously cultured.

### Results:

Virus	Known Specimens (n=44)			Unknown Specimens (n=96)	
	Culture	Antigen	RVP	Culture	RVP
RSV	NT	8	8 (100%)	3	10
Flu A	NT	10	9 (90%)	0	1
Flu B	NT	10	10(100%)	0	0
Ad	8	NT	7 (88%)	6	3
EV/Rh	8	NT	8(100%)	NT	25
CoV	NT	NT	0	NT	2
HMPV	NT	NT	0	NT	5
<b>Total</b>	<b>16</b>	<b>28</b>	<b>42</b>	<b>9</b>	<b>46</b>

### Conclusions:

The Luminex Diagnostics® RVP identified 46 viruses among 96 specimens previously reported as Flu or RSV antigen negative. 6 viruses were detected by RVP and culture while 40 viruses were detected by RVP only. The addition of Luminex Molecular Diagnostics® RVP to the viral testing algorithm of respiratory infections in children may improve diagnostic yield of clinical specimen and provide the opportunity to potentially decrease antibiotic use, laboratory testing, and hospital costs.

## INTRODUCTION

Respiratory viruses are common pathogens that cause clinical disease in children year round. Rapid identification of the respiratory virus in children will improve patient care by reducing the unnecessary use of antibiotics, reduce hospital costs and improve infection control practices. Rapid antigen tests for Flu A/B and RSV provide results within 1 hr and is useful in clinical decision making. However these tests suffer from poor sensitivity and it is imperative to reflex antigen-negative specimens to a confirmatory test for an optimal diagnostic yield. Cell culture and direct fluorescent antibody (DFA) techniques are good confirmatory tests, but suffer from certain limitations. Cell culture is laborious, time consuming and currently only few respiratory viruses can be detected by cell culture. Although DFA is a rapid test with acceptable sensitivity; the test is subjective and results in varied sensitivities based on the expertise of the technologist.

The advent of nucleic acid amplification techniques has provided a more efficient and rapid method of diagnosing respiratory viruses. With the use of multiplex PCR technology one respiratory viral specimen can be tested for many different viruses in a single testing event. Since these tests can be performed within one day a potential impact in the overall care of children with one of these respiratory infections might be realized.

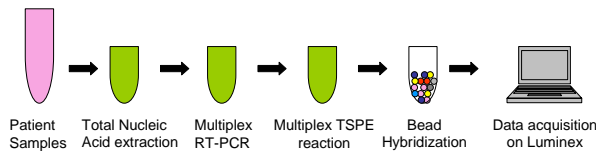
We evaluated the ID-Tag™ respiratory virus panel (RVP) kit that utilizes Luminex technology in detecting the following respiratory viruses; influenza A & B, parainfluenza 1-4, adenovirus, RSV A & B, human metapneumovirus, rhinovirus/enterovirus, and 4 serotypes of coronaviruses. The performance of ID-Tag assay was evaluated with frozen or fresh specimens previously positive for certain viruses by either antigen or culture (Known specimens) and a subset of specimens previously negative for Flu or RSV-antigens (Unknown specimens). These unknown specimen were simultaneously setup for culture and ID-Tag testing.

## MATERIALS & METHODS

**Specimens:** Nasopharyngeal specimens obtained from children seen at Children's Mercy Hospital and Clinics with respiratory symptoms.

**44 Known Specimens:**  
28 known by rapid antigen: 8 RSV, 10 Flu A, 10 Flu B  
16detected by cell culture: 8 Adenovirus, 8 Rhinovirus

**96 Unknown Specimens:**  
These specimens were negative by rapid antigen test for RSV or Flu A and B. At the time of the ID-Tag RVP evaluation these specimens were tested again by cell culture techniques- Rmix shell vial culture.

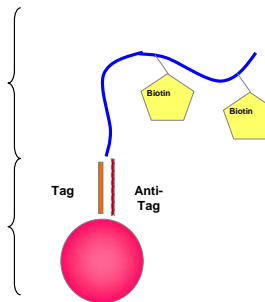


### PCR and TSPE Reaction

Assay designed and optimized so that biotin is incorporated only if the molecular species is present in the clinical sample

### Universal Array Sorting

Oligo-coupled beads from the empirically validated RVM Set used with xTAG products. Use MFI for every target-specific primer to determine presence or absence of each virus



## RESULTS

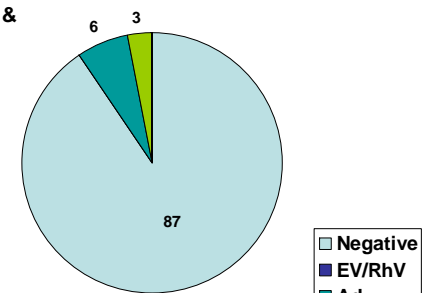
### Known Specimens (n=44)

Virus	Culture	Antigen	RVP ID-Tag
RSV*	NT**	8	8 (100%)
Flu A	NT	10	9 (90%)
Flu B	NT	10	10 (100%)
Adenovirus	8	NT	7 (88%)
Enterovirus/Rhinovirus	8	NT	8 (100%)

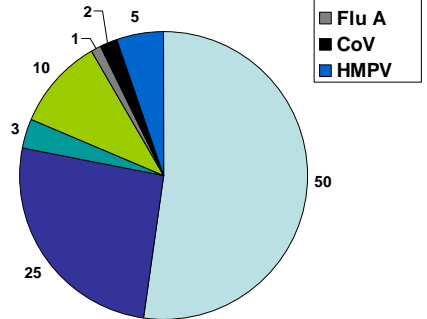
\*Testing was performed on NP specimens obtained during the 2006-07 RSV season  
\*\* Not tested

### Unknown Specimens (n=96)

Rapid antigen & Cell culture



RVP ID-Tag™ with Luminex



- ID-Tag RVP™ detected in 95% (42/44) of known respiratory pathogens previously identified in nasopharyngeal specimens
- ID-Tag RVP™ detected an additional 40 viruses in unknown specimens
  - Enterovirus/Rhinovirus most frequently isolated, 25/96 (26%)
  - HMPV was identified in 5 specimens (5%)
  - A coronavirus was identified in 2 specimens (2%)
- No Co-infection was observed in any of the specimens

## CONCLUSIONS

- ID-Tag RVP™ using Luminex technology is an easy and rapid method to detect respiratory viruses in children.
- The ID-Tag RVP assay increased the diagnostic yield of respiratory viruses from NP specimens by 4 fold.
- Future studies will evaluate the impact of this diagnostic test on length of hospital stay and total antibiotic use in children with acute respiratory viral disease.