

# EVALUATION OF THE DIAGNOSTIC HYBRIDS D<sup>3</sup> *ULTRA DUET*<sup>TM</sup> RESPIRATORY VIRUS IDENTIFICATION KIT FOR THE DETECTION OF RESPIRATORY VIRUSES DIRECTLY FROM CLINICAL SPECIMENS

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**INTRODUCTION:** The rapid detection of respiratory pathogens (Influenza A and B, Parainfluenza 1, 2, and 3, Adenovirus, RSV, and Metapneumovirus) by direct immunofluorescence (DFA) has a significant impact on therapeutic choices and the prevention of nosocomial outbreaks. While rapid tests for Influenza A/B and RSV are convenient testing alternatives during peak seasons, they lack the sensitivity of DFA testing and will not detect Adenovirus, Metapneumovirus, or Parainfluenza virus. Traditional DFA testing requires a wash step, a centrifugation step to pellet the cells, then dotting a slide with a small amount of the cell pellet. The slides must then be dried and fixed before staining. A positive screening DFA using a respiratory virus pool requires additional testing with individual antibodies to identify the specific virus. The D<sup>3</sup> *Ultra Duet* Respiratory Virus Identification Kit (Diagnostic Hybrids [DHI], Athens, OH) allows, by the use of three individual stains each having different fluors, the specific identification of eight respiratory viruses using a ‘wet’ stain. Each stain is specific for two different virus types:

- Blue bottle stain = Influenza A (gold)/ Influenza B (green)
- Orange bottle stain = RSV (gold)/Metapneumovirus (green)
- Yellow bottle stain = Parainfluenza (gold)/ Adenovirus (green)

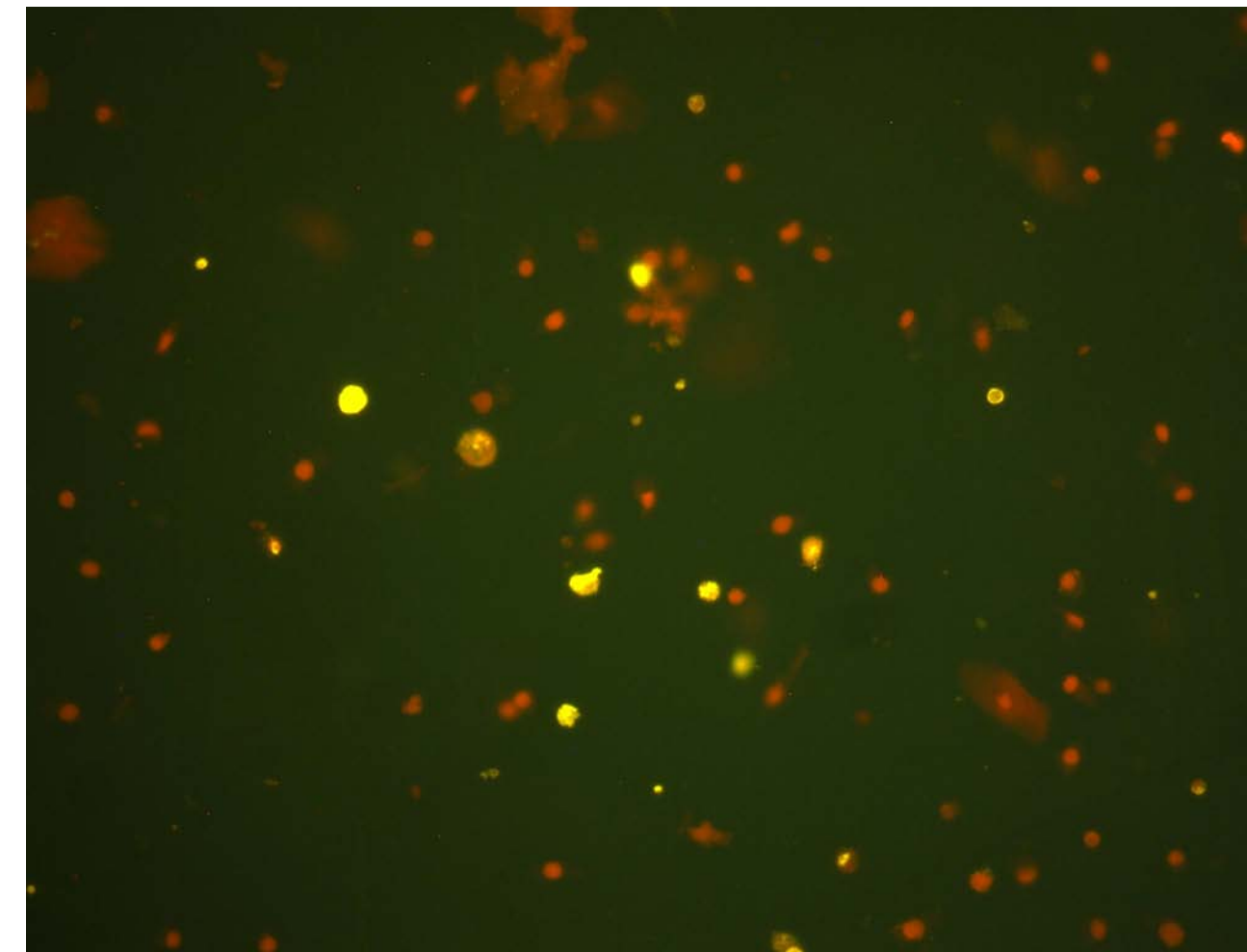
With the wet stain procedure, drying time and fixing time are eliminated. A small amount of the cell pellet is mixed with each of the three stains in separate bullet tubes. Incubation time is shortened to 5 minutes and after a 2 minute wash cycle, the resulting cell button/stain mixture is put directly onto a slide and cover slipped for reading. The purpose of this study was to evaluate the D<sup>3</sup> *Ultra Duet* Respiratory Virus Identification Kit and compare results to the D<sup>3</sup> RSV Duet Respiratory Virus Screening Kit and the Metapneumovirus ID reagent, individual respiratory virus DFA stains and R-Mix<sup>TM</sup> culture.

**MATERIALS AND METHODS:** For the evaluation, 444 specimens (nasal washing [NW], nasopharyngeal swabs [NPS], and nasopharyngeal aspirates [NPA]) were processed according to standard laboratory practices for the preparation of dry slide DFA testing, ‘wet stain’ preparation, viral culture planting and staining on R-Mix vials. ‘Wet stains’ were tested for Influenza A/B, RSV/Metapneumovirus, and Parainfluenza/Adenovirus. Results were compared to dry slide DFA, and R-Mix virus culture. Dry slides were screened using individual Metapneumovirus stain and RSV/respiratory pool stain. All positive screens were then tested using individual respiratory virus stains (unless positive for Metapneumovirus or RSV).

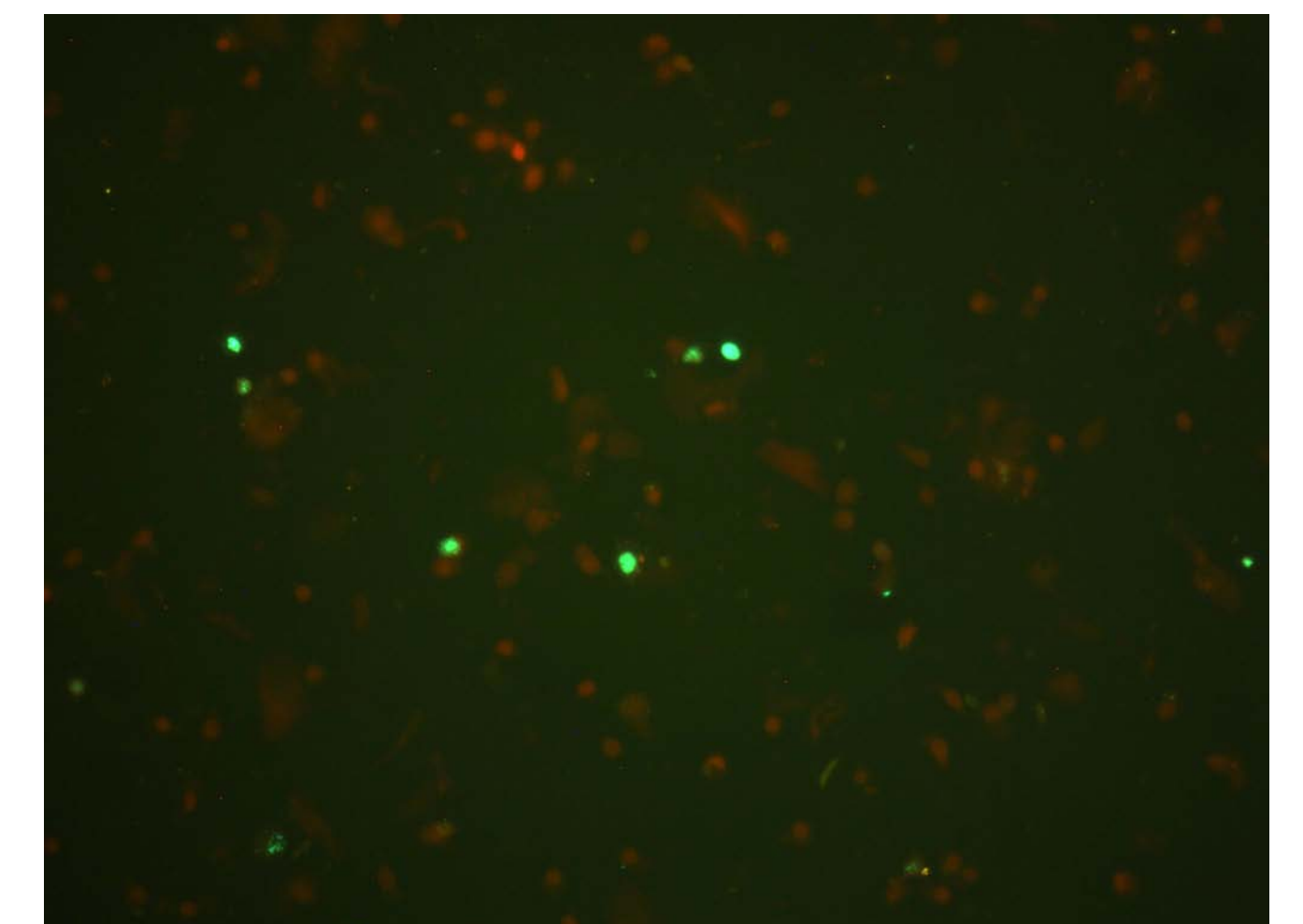
**TABLE 1: Results of for the D<sup>3</sup> Ultra Duet compared to D<sup>3</sup> Ultra RSV DUET DFA and R-Mix Culture (N = 444 )**

Virus	Ultra Duet compared to Ultra DFA		Ultra Duet compared to R-Mix Culture	
	PPA	NPA	Sensitivity	Specificity
Influenza A	100% (39/39)	100% (405/405)	83.7% (36/43)	99.3% (398/401)
Influenza B	100% (5/5)	100% (439/439)	60.0% (3/5)	99.5% (437/439)
RSV	100% (59/59)	100% (385/385)	93.6% (44/47)	96.2% (382/397)
MPV	97.4% (37/38)	100% (406/406)	96.4% (27/28)	97.6% (401/4116)
Parainfluenza	100% (24/24)	100% (420/420)	91.3% (21/23)	99.3% (418/421)
Adenovirus	100% (4/4)	100% (440/440)	75.0% (3/4)	99.8% (439/440)

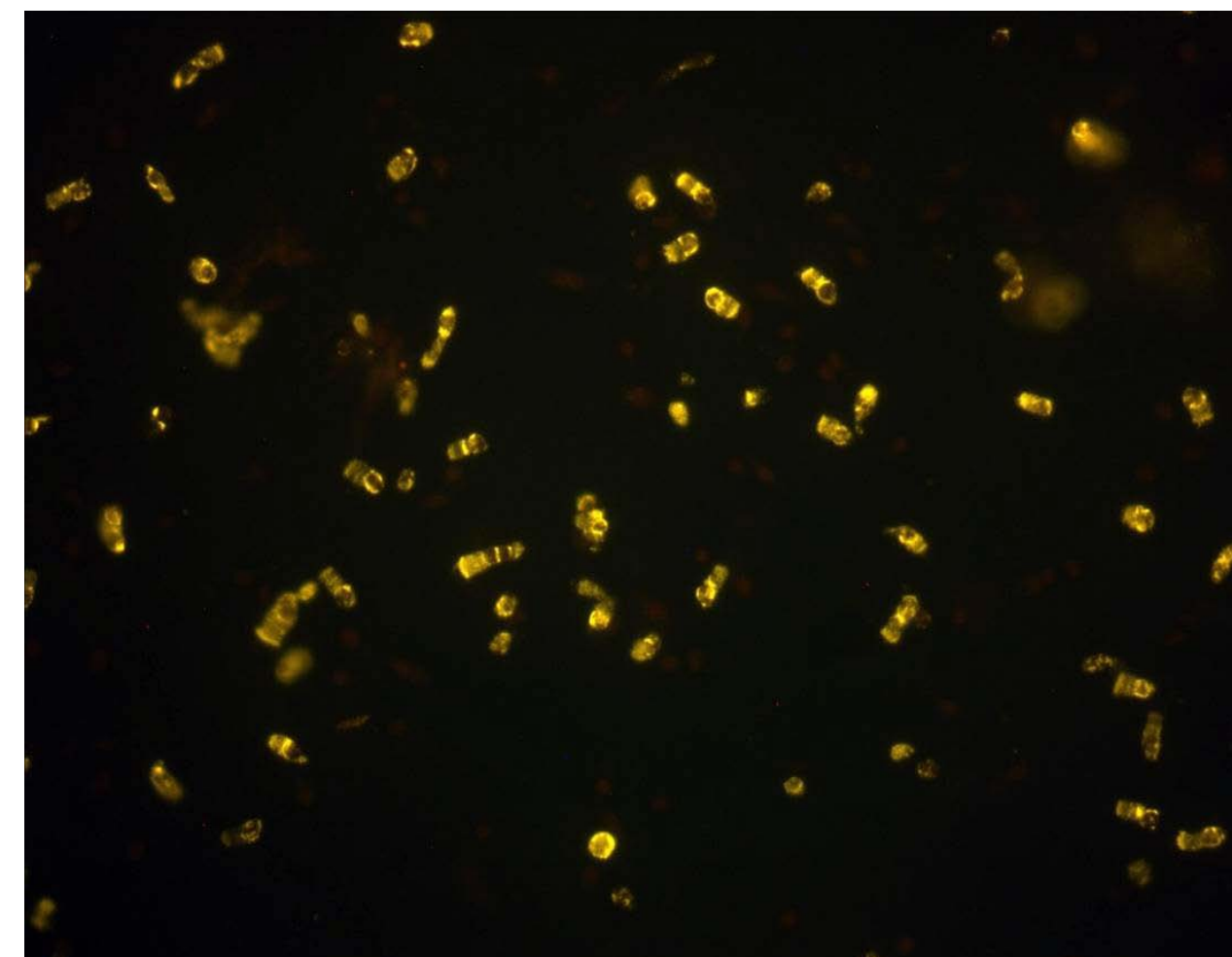
**Influenza A D<sup>3</sup> Ultra Duet positive specimen**



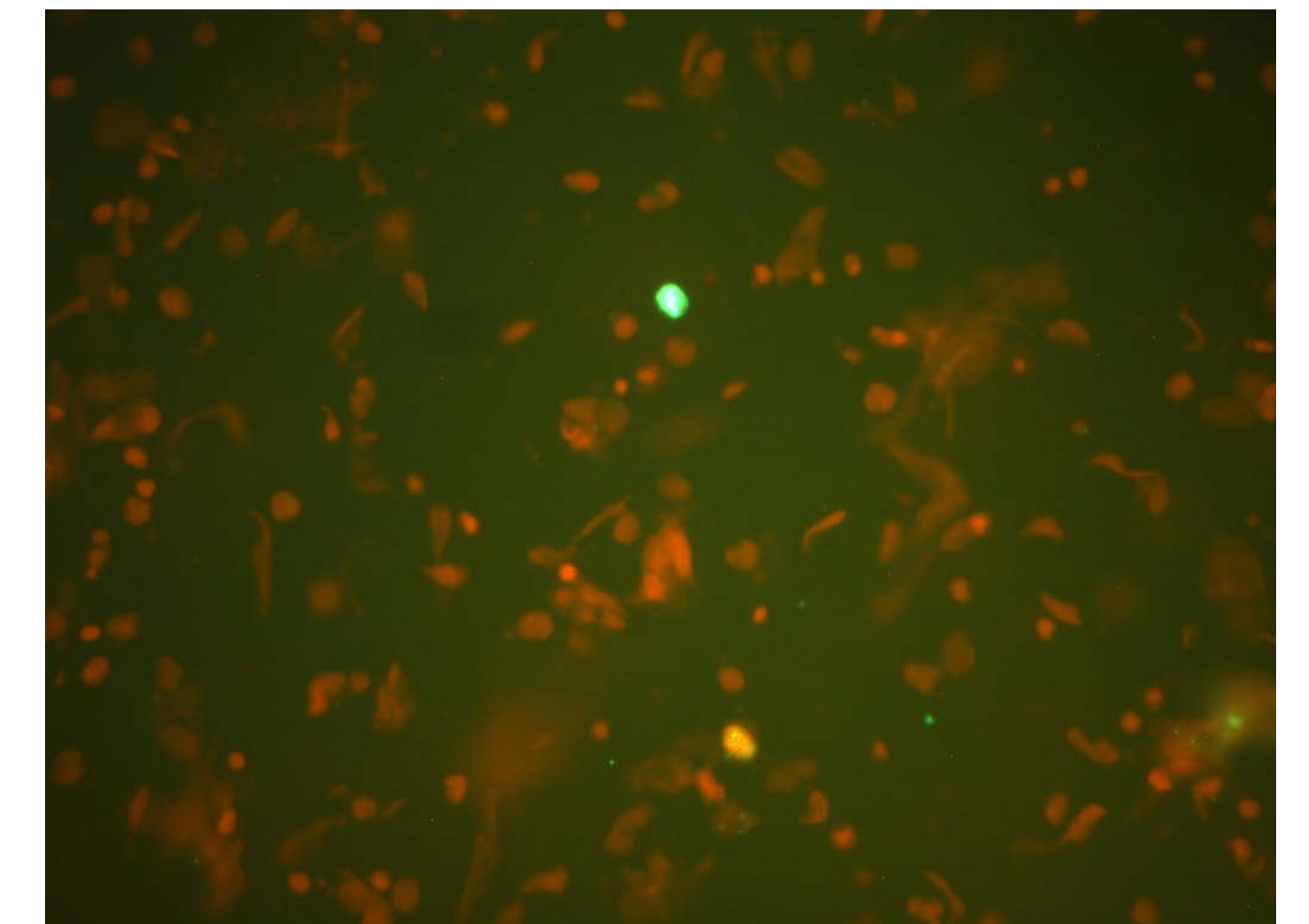
**Influenza B D<sup>3</sup> Ultra Duet positive specimen**



**Parainfluenza D<sup>3</sup> Ultra Duet positive specimen**



**Adenovirus D<sup>3</sup> Ultra Duet positive specimen**



**Conclusion:** The D<sup>3</sup> *Ultra Duet* Respiratory Virus Identification Kit showed comparable sensitivity to traditional DFA testing and slightly lower sensitivity to R-Mix virus culture. The ability to accurately and rapidly detect Influenza A, Influenza B, RSV, Metapneumovirus, Parainfluenza and Adenovirus, specifically within one procedure, can reduce turn around time to results and technical time by not having to test separate slides with individual reagents on respiratory pool positive samples.