Section 05, 510(k) Summary

Applicant:

DIAGNOSTIC HYBRIDS, INC.
1055 East State Street
Suite 100
Athens, OHIO 45701

Contact Information:

Ronald H. Lollar, Senior Director Product Realization, Management and Marketing
1055 East State Street
Suite 100
Athens, Ohio 45701
740-589-3300 – Corporate number
740-589-3373 – Desk phone
740-593-8437 – Fax
lollar@dhiusa.com

Date of preparation of 510(k) summary:

April 17, 2009

Device Name:

Trade name – **D³ FastPoint L-DFA Respiratory Virus Identification Kit**
Common name – Respiratory virus DFA assay
Classification name – Antisera, Cf, Influenza Virus A, B, C
Product Code – GNW
Regulation – 21 CFR 866.3330, Class I, Influenza virus serological reagents; Panel Microbiology (83)

Legally marketed devices to which equivalence is claimed:

D³ *Ultra* DFA Respiratory Virus Screening & ID Kit (k061101)

Intended Use: The Diagnostic Hybrids, Inc. D³ *Ultra* DFA (direct fluorescent antibody) Respiratory Virus Screening & ID Kit (D³ *Ultra*) is intended for the qualitative detection and identification of the influenza A, influenza B, respiratory syncytial virus (RSV), adenovirus, parainfluenza 1, parainfluenza 2 and parainfluenza 3 virus in respiratory specimens, by either direct detection or cell culture method, by immunofluorescence using fluoresceinated monoclonal antibodies (MAbs). It is recommended that specimens found to be negative after examination of the direct specimen

result be confirmed by cell culture. Negative results do not preclude respiratory virus infection and should not be used as the sole basis for diagnosis, treatment or other management decisions.

- Performance characteristics for influenza A were established when influenza A/H3 and A/H1 were the predominant influenza A viruses in circulation. When other influenza A viruses are emerging, performance characteristics may vary.
- If infection with a novel influenza A virus is suspected based on current clinical and epidemiological screening criteria recommended by public health authorities, specimens should be collected with appropriate infection control precautions for novel virulent influenza viruses and sent to state or local health departments for testing. Viral culture should not be attempted in these cases unless a BSL3+ facility is available to receive and culture specimens.

D³ *Duet* DFA RSV/Respiratory Virus Screening Kit (k081928)

The Diagnostic Hybrids, Inc. device, D³ *Duet* DFA RSV/Respiratory Virus Screening Kit (D³ *Duet* RSV Kit), is intended for the qualitative detection and identification of respiratory syncytial virus, while screening for influenza A virus, influenza B virus, adenovirus, and parainfluenza virus types 1, 2 and 3 viral antigens, in nasal and nasopharyngeal swabs and aspirates or in cell culture. The assay detects viral antigens by immunofluorescence using monoclonal antibodies (MAbs), from patients with signs and symptoms of respiratory infection.

It is recommended that specimens found to be negative after examination of the direct specimen result be confirmed by cell culture. Negative results do not preclude influenza virus infection and should not be used as the sole basis for diagnosis, treatment or other management decisions.

Performance characteristics for influenza A virus detection and identification were established when influenza A (H3N2) and influenza A (H1N1) were the predominant influenza A strains circulating in the United States. Performance characteristics for influenza A virus detection and identification were established when influenza A H3N2 and influenza A H1N1 were the predominant influenza A strains circulating in the United States. When other influenza A viruses are emerging, performance characteristics may vary. If infection with a novel influenza A virus is suspected based on current clinical and epidemiological screening criteria recommended by public health authorities, specimens should be collected with appropriate infection control precautions for novel virulent influenza viruses and sent to a state or local health department for testing. Viral culture should not be

attempted in these cases unless a BSL 3+ facility is available to receive and culture specimens.

D³ DFA Metapneumovirus Identification Kit (k090073)

The Diagnostic Hybrids, Inc. device, D³ DFA Metapneumovirus Identification Kit (D³ MPV Kit), is intended for the qualitative detection and identification of human metapneumovirus (hMPV) in nasal and nasopharyngeal swabs and aspirates/washes or cell culture. The assay detects hMPV antigens by immunofluorescence using a blend of three monoclonal antibodies (MAbs), from patients with signs and symptoms of acute respiratory infection. This assay detects but is not intended to differentiate the four recognized genetic sub-lineages of hMPV.

Negative results do not preclude hMPV infection and should not be used as the sole basis for diagnosis, treatment or other management decisions. It is recommended that specimens found to be negative after examination of the direct specimen results be confirmed by an FDA-cleared hMPV molecular assay.

Device Description:

The D³ FastPoint L-DFA Respiratory Virus Identification Kit uses three blends (each called a "L-DFA Reagent") of viral antigen-specific murine monoclonal antibodies that are directly labeled with either R-PE (influenza A virus, respiratory syncytial virus, and parainfluenza virus) or fluorescein (influenza B virus, metapneumovirus, and adenovirus) for the rapid identification of respiratory viruses in nasal and nasopharyngeal swabs and aspirates from patients with signs and symptoms of respiratory infection.

Kit Components:

1. **D³ FastPoint L-DFA Influenza A/Influenza B Reagent**, 4.0-mL. One dropper bottle containing a mixture of PE-labeled murine monoclonal antibodies directed against influenza A virus antigens and FITC-labeled murine monoclonal antibodies directed against influenza B virus antigens. The buffered, stabilized, aqueous solution contains Evans Blue and propidium iodide as counter-stains and 0.1% sodium azide as preservative.

2. **D³ FastPoint L-DFA RSV/MPV Reagent, 4.0-mL**. One dropper bottle containing a mixture of PE-labeled murine monoclonal antibodies directed against respiratory syncytial virus antigens and FITC-labeled murine monoclonal antibodies directed against metapneumovirus antigens. The buffered, stabilized, aqueous solution contains Evans Blue and propidium iodide as counter-stains and 0.1% sodium azide as preservative.

3. **D³ FastPoint L-DFA PIV/Adenovirus Reagent**, 4.0-mL. One dropper bottle containing a mixture of PE-labeled murine monoclonal antibodies directed against parainfluenza virus types 1, 2, or 3 antigens and FITC-labeled murine monoclonal antibodies directed against adenovirus antigens. The buffered, stabilized, aqueous solution contains Evans Blue and propidium iodide as counter-stains and 0.1% sodium azide as preservative.

4. **40X PBS Concentrate, 25-mL**. One bottle of 40X PBS concentrate containing 4% sodium azide (0.1% sodium azide after dilution to 1X using de-mineralized water).

5. **Re-suspension Buffer**, 6.0-mL. One bottle of a buffered glycerol solution and 0.1% sodium azide.

6. **D³ FastPoint L-DFA Respiratory Virus Antigen Control Slides**, 5-slides. Five individually packaged control slides containing 6 wells with cell culture-derived positive and negative control cells. Each positive well is identified as to the virus infected cells present, i.e., influenza A virus, influenza B virus, respiratory syncytial virus, metapneumovirus, parainfluenza virus, and adenovirus. The negative wells contain non-infected cells. Each slide is intended to be stained only one time.

The cells to be tested are derived from respiratory specimens from patients with signs and symptoms of respiratory infection. The cells are permeabilized and stained concurrently in a liquid suspension format in 3 separate vials, each containing one of the 3 above reagents. After incubating at 35°C to 37°C for 5 minutes, the stained cell suspensions are rinsed with 1X PBS. The rinsed cells are pelleted by centrifugation and then re-suspended with the resuspension buffer and loaded onto a specimen slide well. The cells are examined using a fluorescence microscope. Cells infected with influenza A virus, respiratory syncytial virus, or parainfluenza virus types 1, 2 and 3 will exhibit golden-yellow fluorescence due to the PE. Cells infected with influenza B virus, metapnemovirus or adenovirus will exhibit apple-green fluorescence due to the FITC. Non-infected cells will exhibit red fluorescence due to the Evans Blue counter-stain. Nuclei of intact cells will exhibit orange-red fluorescence due to the propidium iodide.

Intended Use:

The Diagnostic Hybrids, Inc. device, D³ FastPoint L-DFA Respiratory Virus Identification Kit is intended for the qualitative identification of influenza A virus, influenza B virus, respiratory syncytial virus, human metapneumovirus, adenovirus and to screen for the presence of parainfluenza virus types 1, 2, and 3 in nasal and nasopharyngeal swabs and aspirates/washes specimens from patients with signs and symptoms of respiratory infection by direct detection of immunofluorescence using monoclonal antibodies (MAbs).

It is recommended that specimens found to be negative for influenza A virus, influenza B virus, respiratory syncytial virus, adenovirus or parainfluenza viruses after examination of the direct specimen result be confirmed by cell culture. Specimens found to be negative for human metapneumovirus after examination of the direct specimen results should be confirmed by an FDA cleared human metapneumovirus molecular assay. Negative results do not preclude respiratory virus infection and should not be used as the sole basis for diagnosis, treatment or other management decisions.

Performance characteristics for influenza A virus detection and identification were established when influenza A (H3N2) and influenza A (H1N1) were the predominant influenza A strains circulating in the United States. Since influenza strains display antigenic drift and shift from year to year, performance characteristics may vary. If infection with a novel influenza A virus is suspected, based on clinical and epidemiological screening criteria communicated by public health authorities, collect specimens following appropriate infection control precautions and submit to state or local health departments, for testing. Viral culture should not be attempted in these cases unless a BSL 3+ facility[1] is available to receive and culture specimens.[2]

Technological Characteristics, Compared to Predicate Device:

Table 5.1: Characteristics of the D³ FastPoint L-DFA Kit are compared to those of the following Diagnostic Hybrids (DHI) predicate devices

Characteristics	D³ FastPoint L-DFA Kit Subject Device	D³ *Ultra* Kit 510(k) #k061101	D³ *Duet* RSV Kit 510(k) # k081928	D³ MPV Kit 510(k) # k090073
Intended Use	The Diagnostic Hybrids, Inc. device, D³ FastPoint L-DFA Respiratory Virus Identification Kit is intended for the	The Diagnostic Hybrids, Inc. D³ *Ultra*™ DFA (direct fluorescent antibody) Respiratory Virus	The Diagnostic Hybrids, Inc. device, D³ Duet DFA RSV/Respiratory Virus Screening Kit, is intended for the	The Diagnostic Hybrids, Inc. device, D³ DFA Metapneumovirus Identification Kit, is intended for the

[1] www.cdc.gov
[2] FDA Guidance Document: In Vitro Diagnostic Devices to Detect Influenza A Viruses: Labeling and Regulatory Path; Issued 4/10/2006

Table 5.1: Characteristics of the D³ FastPoint L-DFA Kit are compared to those of the following Diagnostic Hybrids (DHI) predicate devices

Characteristics	D³ FastPoint L-DFA Kit Subject Device	D³ *Ultra* Kit 510(k) #k061101	D³ *Duet* RSV Kit 510(k) # k081928	D³ MPV Kit 510(k) # k090073
	qualitative identification of influenza A virus, influenza B virus, respiratory syncytial virus, human metapneumovirus, adenovirus and to screen for the presence of parainfluenza virus types 1, 2, and 3 in nasal and nasopharyngeal swabs and aspirates/washes specimens from patients with signs and symptoms of respiratory infection by direct detection of immunofluorescence using monoclonal antibodies (MAbs). It is recommended that specimens found to be negative for influenza A virus, influenza B virus, respiratory syncytial virus, adenovirus or parainfluenza viruses after examination of the direct specimen result be confirmed by cell culture. Specimens found to be negative for human metapneumovirus after examination of the direct specimen results should be confirmed by an FDA cleared human	Screening & ID Kit is intended for the qualitative detection and identification of the influenza A, influenza B, respiratory syncytial virus (RSV), adenovirus, parainfluenza 1, parainfluenza 2 and parainfluenza 3 virus in respiratory specimens, by either direct detection or cell culture method, by immunofluorescence using monoclonal antibodies (MAbs). It is recommended that specimens found to be negative after examination of the direct specimen result be confirmed by cell culture. Negative results do not preclude respiratory virus infection and should not be used as the sole basis for diagnosis, treatment or other management decisions.	qualitative detection and identification of respiratory syncytial virus, while screening for influenza A virus, influenza B virus, adenovirus, and parainfluenza virus types 1, 2 and 3 viral antigens, in nasal and nasopharyngeal swabs and aspirates or in cell culture. The assay detects viral antigens by immunofluorescence using monoclonal antibodies (MAbs), from patients with signs and symptoms of respiratory infection. It is recommended that specimens found to be negative after examination of the direct specimen result be confirmed by cell culture. Negative results do not preclude influenza virus infection and should not be used as the sole basis for diagnosis, treatment or other management decisions.	qualitative detection and identification of human metapneumovirus (hMPV) in nasal and nasopharyngeal swabs and aspirates/washes or cell culture. The assay detects hMPV antigens by immunofluorescence using a blend of three monoclonal antibodies (MAbs), from patients with signs and symptoms of acute respiratory infection. This assay detects but is not intended to differentiate the four recognized genetic sub-lineages of hMPV. Negative results do not preclude hMPV infection and should not be used as the sole basis for diagnosis, treatment or other management decisions. It is recommended that specimens found to be negative after examination of the direct specimen results be confirmed by an FDA-cleared hMPV molecular assay.

Table 5.1: Characteristics of the D³ FastPoint L-DFA Kit are compared to those of the following Diagnostic Hybrids (DHI) predicate devices

Characteristics	D³ FastPoint L-DFA Kit Subject Device	D³ *Ultra* Kit 510(k) #k061101	D³ *Duet* RSV Kit 510(k) # k081928	D³ MPV Kit 510(k) # k090073
	metapneumovirus molecular assay. Negative results do not preclude respiratory virus infection and should not be used as the sole basis for diagnosis, treatment or other management decisions.			
Target Viruses	influenza A virus, influenza B virus, respiratory syncytial virus, metapneumovirus, adenovirus, parainfluenza virus type 1, parainfluenza virus type 2, parainfluenza virus type 3	influenza A virus, influenza B virus, respiratory syncytial virus, adenovirus, parainfluenza virus type 1, parainfluenza virus type 2, parainfluenza virus type 3	influenza A virus, influenza B virus, respiratory syncytial virus, adenovirus, parainfluenza virus type 1, parainfluenza virus type 2, parainfluenza virus type 3	metapneumovirus
Monoclonal antibodies (MAbs)	The D³ FastPoint L-DFA Reagents contain 18 MAbs to 8 different respiratory viruses (influenza A virus, influenza B virus, respiratory syncytial virus, metapneumovirus, adenovirus, parainfluenza virus type 1, parainfluenza virus type 2, parainfluenza virus type 3)	The Respiratory Virus DFA Screening Reagent contains 15 MAbs to 7 different respiratory viruses (influenza A virus, influenza B virus, respiratory syncytial virus, adenovirus, parainfluenza virus type 1, parainfluenza virus type 2, parainfluenza virus type 3)	The RSV/Respiratory Virus DFA Screening Reagent contains 15 MAbs to 7 different respiratory viruses (influenza A virus, influenza B virus, adenovirus, parainfluenza virus type 1, parainfluenza virus type 2, parainfluenza virus type 3), plus 2 MAbs to respiratory syncytial virus.	The Metapneumovirus DFA Reagent contains 3 MAbs to metapneumovirus
Labeling method	Direct labeling, - using R-Phycoerythrin (R-PE) to label the MAbs to influenza	Direct labeling,	Direct labeling, - using R-Phycoerythrin (R-PE) to label the MAbs to respiratory	Direct labeling,

Table 5.1: Characteristics of the D³ FastPoint L-DFA Kit are compared to those of the following Diagnostic Hybrids (DHI) predicate devices

Characteristics	D³ FastPoint L-DFA Kit Subject Device	D³ *Ultra* Kit 510(k) #k061101	D³ *Duet* RSV Kit 510(k) # k081928	D³ MPV Kit 510(k) # k090073
	A virus, RSV and parainfluenza virus types 1, 2 and 3. - using fluorescein isothiocyanate (FITC) to label influenza B virus, metapneumovirus and adenovirus MAbs with fluorescein.	- using fluorescein isothiocyanate (FITC) to label all MAbs with fluorescein.	syncytial virus. - using fluorescein isothiocyanate (FITC) to label all other MAbs with fluorescein.	- using fluorescein isothiocyanate (FITC) to label all MAbs with fluorescein.
R-Phycoerythrin-labeled MAbs	influenza A virus, respiratory syncytial virus, parainfluenza virus type 1, parainfluenza virus type 2, parainfluenza virus type 3	None	respiratory syncytial virus	None
Fluorescein-labeled MAbs	influenza B virus, metapneumovirus, adenovirus	influenza A virus, influenza B virus, respiratory syncytial virus, adenovirus, parainfluenza virus type 1, parainfluenza virus type 2, parainfluenza virus type 3	influenza A virus, influenza B virus, adenovirus, parainfluenza virus type 1, parainfluenza virus type 2, parainfluenza virus type 3	metapneumovirus
Cell Fixative	Proprietary Non-Acetone based system	Acetone	Acetone	Acetone
Cell Counter-stain	Propidium Iodide, Evans Blue	Evans Blue	Evans Blue	Evans Blue
Performance characteristics				
Staining patterns	**Influenza A and B:** The fluorescence is cytoplasmic or bright nuclear or both. Cells appear round. **Respiratory Syncytial Virus:** The fluorescence is cytoplasmic. Cells appear round. **Metapneumovirus:** The fluorescence is	**Influenza A and B:** The fluorescence is cytoplasmic, nuclear or both. Cytoplasmic staining is often punctate with large inclusions while nuclear staining is uniformly bright. **Respiratory Syncytial Virus:** The fluorescence is	**Influenza A and B:** The fluorescence is cytoplasmic, nuclear or both. Cytoplasmic staining is often punctate with large inclusions while nuclear staining is uniformly bright. **Respiratory Syncytial Virus:**	**Metapneumovirus:** The fluorescence is cytoplasmic and punctate with small inclusions in the syncytia. **Negative:** Entire cell fluoresce red due to the Evans Blue counter-stain.

Table 5.1: Characteristics of the D³ FastPoint L-DFA Kit are compared to those of the following Diagnostic Hybrids (DHI) predicate devices

Characteristics	D³ FastPoint L-DFA Kit Subject Device	D³ *Ultra* Kit 510(k) #k061101	D³ *Duet* RSV Kit 510(k) # k081928	D³ MPV Kit 510(k) # k090073
	cytoplasmic and punctate. Cells appear round. **Parainfluenza 1, 2, 3:** The fluorescence is cytoplasmic. Cells appear round. **Adenovirus:** The fluorescence is cytoplasmic or bright nuclear or both. Cells appear round. **Negative:** Cells fluoresce red due to the Evans Blue counter-stain. **Nuclei:** Cell Nuclei fluoresce orange-red due to the Propidium Iodide counter-stain.	cytoplasmic and punctate with small inclusions in the syncytia. **Parainfluenza 1, 2, 3:** The fluorescence is cytoplasmic and punctate with irregular inclusions. Types 2 and 3 cause the formation of syncytia. **Adenovirus:** The fluorescence is cytoplasmic and punctate or bright nuclear or both. **Negative:** Cells fluoresce red due to the Evans Blue counter-stain.	The fluorescence is cytoplasmic and punctate with small inclusions in the syncytia. **Parainfluenza 1, 2, 3:** The fluorescence is cytoplasmic and punctate with irregular inclusions. Types 2 and 3 cause the formation of syncytia. **Adenovirus:** The fluorescence is cytoplasmic and punctate or bright nuclear or both. **Negative:** Cells fluoresce red due to the Evans Blue counter-stain.	
Analytical specificity (for influenza A virus strains; MAbs are reactive with all listed strains)	13 influenza A strains: Influenza A Mexico/4108/2009 (H1N1) from CDC*, Influenza A California/07/2009 (H1N1) from CDC*, Aichi (H3N2), Mal (H1N1), Hong Kong (H3N2), Denver (H1N1), Port Chalmers (H3N2), Victoria (H3N2), New Jersey (H1N1), WS (H1N1), PR (H1N1), Wisconsin (H3N2), WS (H1N1), A/NWS/33 (H1N1)	10 influenza A strains: Aichi (H3N2), Mal (H1N1), Hong Kong (H3N2), Denver (H1N1), Port Chalmers (H3N2), Victoria (H3N2), New Jersey (H1N1), WS (H1N1), PR,(H1N1), A/NWS/33 (H1N1)	10 influenza A strains: Aichi (H3N2), Mal (H1N1), Hong Kong (H3N2), Denver (H1N1), Port Chalmers (H3N2), Victoria (H3N2), New Jersey (H1N1), WS (H1N1), PR (H1N1), A/NWS/33 (H1N1)	No reaction was seen to any of the tested influenza A viruses with the Metapneumovirus DFA Reagent
Analytical specificity (for Influenza B virus strains; MAbs are reactive with all listed strains)	7 influenza B strains: Hong Kong, Maryland, Mass,	7 influenza B strains: Hong Kong, Maryland, Mass,	7 influenza B strains: Hong Kong, Maryland, Mass,	No reaction was seen to any of the tested influenza B

Table 5.1: Characteristics of the D³ FastPoint L-DFA Kit are compared to those of the following Diagnostic Hybrids (DHI) predicate devices					
Characteristics		D³ FastPoint L-DFA Kit Subject Device	D³ *Ultra* Kit 510(k) #k061101	D³ *Duet* RSV Kit 510(k) # k081928	D³ MPV Kit 510(k) # k090073
		GL, Taiwan, B/Lee/40, Russia	GL, Taiwan, B/Lee/40, Russia	GL, Taiwan, B/Lee/40, Russia	viruses with the Metapneumovirus DFA Reagent
Analytical specificity (cross-reactivity studies; various strains of microorganisms and cell lines)	Viruses	22	31	32	59
	Bacteria	22	18	25	25
	Chlamydia spp.	1	1	3	3
	Yeast	1	0	1	1
	Protozoan	01	0	1	1
	Cell lines	N/A	17	17	16

*Although the D³ FastPoint L-DFA Influenza A/Influenza B Reagent has been shown to detect the 2009 H1N1 virus in two culture isolates, the performance characteristics of this device with clinical specimens that are positive for the 2009 H1N1 influenza virus have not been established. The D³ FastPoint L-DFA Influenza A/Influenza B DFA Reagent can distinguish between influenza A and B viruses, but it cannot differentiate influenza subtypes.

Analytical Performance:

Precision/Reproducibility:

Assay precision, intra-assay variability and inter assay variability were assessed with 3 panels of proficiency-level antigen control slides. Each of the 3 reproducibility panels consisted of 5 randomized panel members.

The Influenza A/B panel consisted of the following:

a. Low level influenza A (Victoria strain) infected cells.
b. Low level influenza B (Taiwan strain) infected cells.
c. Low level influenza A (Victoria strain) infected cells mixed with mid level influenza B (Taiwan strain) infected cells.
d. Low level influenza B (Victoria strain) infected cells mixed with mid level influenza A (Victoria strain) infected cells.
e. Mid level non-infected (negative) cells.

The RSV/hMPV panel consisted of the following:

a. Low level RSV (Washington strain) infected cells.
b. Low level hMPV (A1 subtype) infected cells.
c. Low level RSV (Washington strain) infected cells mixed with mid level hMPV (A1 subtype) infected cells.
d. Low level hMPV (A1 subtype) infected cells mixed with mid level RSV (Washington strain) infected cells.
e. Mid level non-infected (negative) cells.

The HPIV/Adenovirus panel consisted of the following:

a. Low level Para 1 (C-35 strain) infected cells.
b. Low level Adenovirus (ATCC type 1) infected cells.
c. Low level Para 1 (C-35 strain) infected cells mixed with mid level Adenovirus (ATCC type 1) infected cells.
d. Low Adenovirus (ATCC type 1) infected cells mixed with mid level Para 1 (C-35 strain) infected cells.
e. Mid level non-infected (negative) cells.

The low level is estimated to contain between 4 to 10% infected cells in the sample. The mid level is estimated to contain between 20 to 25% infected cells in the sample. Each sample contains 2.5 $x10^5$ to 3.5 $x10^5$ total cells.

Each panel was tested daily in two separate runs for 5-days by four different laboratories (40 total runs). The following results were recorded:

a. Presence or absence of golden-yellow fluorescence.
b. Percent of cells exhibiting golden-yellow fluorescence.
c. Presence or absence of apple-green fluorescence.
d. Percent of cells exhibiting apple-green fluorescence.

For the D³ FastPoint L-DFAInfluenza A/Influenza B Reagent, the combined data from the four Study Sites demonstrated reproducible detection of influenza A virus by the R-PE labeled MAbs and reproducible detection of influenza B virus by the FITC-labeled MAbs. The presence of influenza A virus infected cells was reported in 100% (120/120) of the wells in which the infected cells were expected. The presence of influenza B virus infected cells was reported in 100% (120/120) of the wells in which the infected cells were expected. The absence of infected cells was reported in 95% (38/40) of the wells in which infected cells were not present. The total percent agreement for the D³ FastPoint L-DFAa A/Influenza B Reagent was 99.3% (278/280):

Table 5.2: Reproducibility Study Results using the D³ FastPoint L-DFA Influenza A/Influenza B Reagent

	Panel Member	Negative	Flu A Low Level	Flu B Low Level	Mixed Infection		Mixed Infection		Total % Agreement
					Flu A Mid Level	Flu B Low Level	Flu A Low Level	Flu B Mid Level	
	Concentration	No infected cells	4 to 10% infected cells	4 to 10% infected cells	20 to 30% infected cells	4 to 10% infected cells	4 to 10% infected cells	20 to 30% infected cells	
Site 1	Agreement with Expected result	8/10 (80%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	**68/70 (97.1%)**

Table 5.2: Reproducibility Study Results using the D³ FastPoint L-DFA Influenza A/Influenza B Reagent									
Site 2	Agreement with Expected result	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	**70/70 (100%)**
Site 3	Agreement with Expected result	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	**70/70 (100%)**
Site 4	Agreement with Expected result	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	**70/70 (100%)**
	Total Agreement with Expected result	38/40 (95%)	40/40 (100%)	40/40 (100%)	40/40 (100%)	40/40 (100%)	40/40 (100%)	40/40 (100%)	**278/280 (99.3%)**
	95% CI	83.1 – 99.4%	91.2 – 100%	91.2 – 100%	91.2 – 100%	91.2 – 100%	91.2 – 100%	91.2 – 100%	**97.4 – 99.9%**

For the D³ FastPoint L-DFA RSV/hMPV Reagent, the combined data from the four Study Sites demonstrated reproducible detection of RSV by the R-PE labeled MAbs and reproducible detection of hMPV by the FITC-labeled MAbs. The presence of RSV infected cells was reported in 100% (120/120) of the wells in which the infected cells were expected. The presence of hMPV infected cells was reported in 100% (120/120) of the wells in which the infected cells were expected. The absence of infected cells was reported in 100% (40/40) of the wells in which infected cells were not present. The total percent agreement for the D³ FastPoint L-DFA RSV/hMPV Reagent was 100% (280/280):

Table 5.3: Reproducibility Study Results using the D³ FastPoint L-DFA RSV/hMPV Reagent									
	Panel Member	Negative	RSV Low Level	hMPV Low Level	Mixed Infection		Mixed Infection		Total % Agreement
					RSV Mid Level	hMPV Low Level	RSV Low Level	hMPV Mid Level	
	Concentration	No infected cells	4 to 10% infected cells	4 to 10% infected cells	20 to 30% infected cells	4 to 10% infected cells	4 to 10% infected cells	20 to 30% infected cells	
Site 1	Agreement with Expected result	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	**70/70 (100%)**
Site 2	Agreement with Expected result	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	**70/70 (100%)**
Site 3	Agreement with Expected result	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	**70/70 (100%)**

Table 5.3: Reproducibility Study Results using the D³ FastPoint L-DFA RSV/hMPV Reagent									
Site 4	Agreement with Expected result	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	**70/70 (100%)**
	Total Agreement with Expected result	40/40 (100%)	40/40 (100%)	40/40 (100%)	40/40 (100%)	40/40 (100%)	40/40 (100%)	40/40 (100%)	**280/280 (100%)**
	95% CI	91.2 – 100%	91.2 – 100%	91.2 – 100%	91.2 – 100%	91.2 – 100%	91.2 – 100%	91.2 – 100%	**98.7 – 100%**

For the D³ FastPoint L-DFA HPIV/Adenovirus Reagent, the combined data from the four Study Sites demonstrated reproducible detection of HPIV-1 by the R-PE labeled MAbs and reproducible detection of Adenovirus by the FITC-labeled MAbs. The presence of HPIV-1 infected cells was reported in 100% (120/120) of the wells in which the infected cells were expected. The presence of Adenovirus infected cells was reported in 100% (120/120) of the wells in which the infected cells were expected. The absence of infected cells was reported in 100% (40/40) of the wells in which infected cells were not present. The total percent agreement for the D³ FastPoint L-DFA HPIV/Adenovirus was 100% (280/280):

Table 5.4: Reproducibility Study Results using the D³ FastPoint L-DFA HPIV/Adenovirus Reagent									
	Panel Member	Negative	HPIV-1 Low Level	Adenovirus Low Level	Mixed Infection		Mixed Infection		Total % Agreement
					HPIV-1 Mid Level	Adenovirus Low Level	HPIV-1 Low Level	Adenovirus Mid Level	
	Concentration	No infected cells	4 to 10% infected cells	4 to 10% infected cells	20 to 30% infected cells	4 to 10% infected cells	4 to 10% infected cells	20 to 30% infected cells	
Site 1	Agreement with Expected result	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	**70/70 (100%)**
Site 2	Agreement with Expected result	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	**70/70 (100%)**
Site 3	Agreement with Expected result	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	**70/70 (100%)**
Site 4	Agreement with Expected result	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	10/10 (100%)	**70/70 (100%)**
	Total Agreement with Expected result	40/40 (100%)	40/40 (100%)	40/40 (100%)	40/40 (100%)	40/40 (100%)	40/40 (100%)	40/40 (100%)	**280/280 (100%)**

Table 5.4: Reproducibility Study Results using the D³ FastPoint L-DFA HPIV/Adenovirus Reagent									
	95% CI	91.2 – 100%	91.2 – 100%	91.2 – 100%	91.2 – 100%	91.2 – 100%	91.2 – 100%	91.2 – 100%	**98.7 – 100%**

Limit of Detection

Analytical Limit of Detections (LoDs) of the D³ FastPoint L-DFA Reagents was addressed using dilution series of infected model cells. Model cells for 8 characterized respiratory virus isolates; influenza A virus (ATCC Victoria strain), influenza B virus (ATCC Taiwan strain), respiratory syncytial virus (ATCC Washington strain), adenovirus (ATCC type 1), human metapneumovirus subtype A1 (clinical strain), parainfluenza virus types 1, 2, and 3 (ATCC strains C-35, Greer, and C243 respectively) were diluted with non-infected cells to produce a suspension equivalent to 1,000 infected cells per milliliter. This level theoretically yields approximately 25 infected cells per 25-µL of suspension. This suspension was then serially diluted to a theoretical level of less than 1 cell per milliliter. (NOTE: This level was the target to begin with a low positive level. Actual starting levels vary, however, and are within 1 dilution of the 25 infected cell target level). 25-µL aliquots from each dilution level were spotted onto 10 replicate microscope slides, and then stained according to the instructions for use described in this product insert. Each cell spot was examined at 200x magnification. Results were reported as numbers of positive replicates for each set of 10. Analytical detection limits for each of the 8 analytes were defined as the lowest dilutions at which at least 9 out of 10 replicates were detected. LoD study results are summarized in Table 5.5 below:

Table 5.5: Limit of Detections of the D³ FastPoint L-DFA Respiratory Virus Identification Kit			
Virus Strain	Infected cells/mL	Number of replicates with positive cells	LOD determination
Flu A (ATCC Victoria strain)	500	10/10	50 infected cells/mL
	100	10/10	
	50	**10/10**	
	25	5/10	
	12.5	3/10	
	6	2/10	
	3	0/10	
	1.5	2/10	
	0.8	0/10	
	0.4	0/10	
Flu B (ATCC Taiwan strain)	2000	10/10	50 infected cells/mL
	400	10/10	
	200	10/10	
	100	10/10	
	50	**10/10**	
	25	7/10	
	12.5	4/10	
	6	2/10	
	3	0/10	
	1.5	0/10	

Table 5.5: Limit of Detections of the D³ FastPoint L-DFA Respiratory Virus Identification Kit

Virus Strain	Infected cells/mL	Number of replicates with positive cells	LOD determination
RSV (ATCC Washington strain)	1000	10/10	100 infected cells/mL
	200	10/10	
	100	**10/10**	
	50	7/10	
	25	7/10	
	12.5	6/10	
	6	1/10	
	3	0/10	
	1.5	0/10	
	0.8	0/10	
hMPV A1 (Clinical strain)	2000	10/10	100 infected cells/mL
	400	10/10	
	200	10/10	
	100	**10/10**	
	50	6/10	
	25	2/10	
	12.5	0/10	
	6	0/10	
	3	0/10	
	1.5	0/10	
Adenovirus (ATCC type 1)	1000	10/10	100 infected cells/mL
	200	10/10	
	100	**9/10**	
	50	5/10	
	25	1/10	
	12.5	0/10	
	6	0/10	
	3	0/10	
	1.5	0/10	
	0.8	0/10	
HPIV-1 (ATCC strain C-35)	500	10/10	100 infected cells/mL
	100	**10/10**	
	50	6/10	
	25	2/10	
	12.5	1/10	
	6	0/10	
	3	0/10	
	1.5	0/10	
	0.8	0/10	
	0.4	0/10	
HPIV-2 (ATCC strain Greer)	500	10/10	25 infected cells/mL
	100	10/10	
	50	10/10	
	25	**9/10**	
	12.5	6/10	
	6	5/10	
	3	3/10	
	1.5	1/10	
	0.8	0/10	
	0.4	0/10	
HPIV-3 (ATCC strain C243)	1000	10/10	50 infected cells/mL
	200	10/10	
	100	10/10	
	50	**9/10**	

Table 5.5: Limit of Detections of the D³ FastPoint L-DFA Respiratory Virus Identification Kit			
Virus Strain	Infected cells/mL	Number of replicates with positive cells	LOD determination
	25	6/10	
	12.5	2/10	
	6	0/10	
	3	0/10	
	1.5	0/10	
	0.8	0/10	

Analytical reactivity (inclusivity)

Analytical reactivity (inclusivity) of the D³FastPoint L-DFA Influenza A/Influenza B Reagent was evaluated using 13 influenza A virus and 7 influenza B virus strains. Low concentration infected cell suspensions (approximately 4% cells infected, 25-50 infected cells) were prepared for each viral strain. The suspensions were stained with the D³ FastPoint L-DFA Influenza A/Influenza B Reagent.

Table 5.6: Analytical Reactivity (inclusivity) of the D³ FastPoint L-DFA Influenza A/Influenza B Reagent on various influenza A virus and influenza B virus strains		
Influenza Strains	**Infected Cell Concentration (as multiples of the respective established LoD concentration**	**D³ FastPoint L-DFA Influenza A/ Influenza B Reagent Results**
Influenza A Mexico/4108/2009 (H1N1) from CDC*	20x LoD	19 Golden-yellow fluorescent cells
Influenza A California/07/2009 (H1N1) from CDC*	20x LoD	26 Golden-yellow fluorescent cells
Influenza A Wisconsin/56/2005 (H3N2)	20x LoD	39 Golden-yellow fluorescent cells
Influenza A WS, VR-1520 (H1N1)	20x LoD	67 Golden-yellow fluorescent cells
Influenza A Hong Kong, VR-544 (H3N2)	20x LoD	13 Golden-yellow fluorescent cells
Influenza A New Jersey, VR-897 (H1N1)	20x LoD	15 Golden-yellow fluorescent cells
Influenza A A/NWS/33 (H1N1)	20x LoD	10 Golden-yellow fluorescent cells
Influenza A Victoria, VR-822 (H3N2)	20x LoD	10 Golden-yellow fluorescent cells
Influenza A PR, VR-95 (H1N1)	20x LoD	20 Golden-yellow fluorescent cells
Influenza A Port Chalmers, VR-810 (H3N2)	20x LoD	8 Golden-yellow fluorescent cells
Influenza A Aichi, VR-547 (H3N2)	20x LoD	28 Golden-yellow fluorescent cells
Influenza A Denver, VR-546 (H1N1)	20x LoD	30 Golden-yellow fluorescent cells
Influenza A Mal, VR-98 (H1N1)	20x LoD	21 Golden-yellow fluorescent cells
Influenza B GL/1739/54, VR-103	20x LoD	13 Apple-green fluorescent cells
Influenza B Taiwan/2/62, VR-295	20x LoD	44 Apple-green fluorescent cells
Influenza B Hong Kong/5/72, VR-823	20x LoD	21 Apple-green fluorescent cells
Influenza B Maryland/1/59, VR-296	20x LoD	22 Apple-green fluorescent cells
Influenza B Russia, VR-790	20x LoD	36 Apple-green fluorescent cells

Table 5.6: Analytical Reactivity (inclusivity) of the D³ FastPoint L-DFA Influenza A/Influenza B Reagent on various influenza A virus and influenza B virus strains		
Influenza Strains	**Infected Cell Concentration (as multiples of the respective established LoD concentration**	**D³ FastPoint L-DFA Influenza A/ Influenza B Reagent Results**
Influenza B B/Lee/40	20x LoD	41 Apple-green fluorescent cells
Influenza B Massachusetts, VR-523	20x LoD	67 Apple-green fluorescent cells

*Although the D³ FastPoint L-DFA Influenza A/Influenza B Reagent has been shown to detect the 2009 H1N1 virus in two culture isolates, the performance characteristics of this device with clinical specimens that are positive for the 2009 H1N1 influenza virus have not been established. The D³ FastPoint L-DFA Influenza A/Influenza B DFA Reagent can distinguish between influenza A and B viruses, but it cannot differentiate influenza subtypes.

Analytical reactivity (inclusivity) of the D³ FastPoint L-DFA RSV/hMPV DFA Reagent was evaluated using 3 RSV virus and 4 hMPV virus strains. Low concentration infected cell suspensions (approximately 4% cells infected, 25-50 infected cells) were prepared for each viral strain. The suspensions were stained with the D³ FastPoint L-DFA RSV/hMPV Reagent.

Table 5.7: Analytical Reactivity (inclusivity) of the D³ FastPoint L-DFA RSV/hMPV DFA Reagent on various RSV virus and hMPV virus strains		
RSV and hMPV Strains	**Infected Cell Concentration (as multiples of the respective established LoD concentration**	**D³ FastPoint L-DFA RSV/hMPV Reagent Results**
RSV 9320	10x LoD	22 Golden-yellow fluorescent cells
RSV Washington	10x LoD	22 Golden-yellow fluorescent cells
RSV Long	10x LoD	32 Golden-yellow fluorescent cells
hMPV A1	10x LoD	25 Apple-green fluorescent cells
hMPV A2	10x LoD	25 Apple-green fluorescent cells
hMPV B1	10x LoD	25 Apple-green fluorescent cells
hMPV B2	10x LoD	37 Apple-green fluorescent cells

Analytical reactivity (inclusivity) of the D³ FastPoint L-DFA HPIV/Adenovirus DFA Reagent was evaluated using 3 HPIV virus and 10 Adenovirus strains. Low concentration infected cell suspensions (approximately 4% cells infected, 25-50 infected cells) were prepared for each viral strain. The suspensions were stained with the D³ FastPoint L-DFA HPIV/Adenovirus Reagent.

Table 5.8: Analytical Reactivity (inclusivity) of the D³ FastPoint L-DFA HPIV/Adenovirus Reagent on various HPIV virus and adenovirus strains		
Parainfluenza and Adenovirus Strains	**Infected Cell Concentration (as multiples of the respective established LoD concentration**	**D³ FastPoint L-DFA HPIV/Adenovirus Reagent Results**
Parainfluenza 1 C-35	10x LoD	9 Golden-yellow fluorescent cells
Parainfluenza 2 Greer	10x LoD	11 Golden-yellow fluorescent cells
Parainfluenza 3 C-243	10x LoD	22 Golden-yellow fluorescent cells
Adenovirus 1 VR-1	10x LoD	26 Apple-green fluorescent cells
Adenovirus 3 VR-3	10x LoD	17 Apple-green fluorescent cells
Adenovirus 5 VR-5	10x LoD	15 Apple-green fluorescent cells
Adenovirus 6 VR-6	10x LoD	22 Apple-green fluorescent cells
Adenovirus 7 VR-7	10x LoD	16 Apple-green fluorescent cells
Adenovirus 8 VR-1366	10x LoD	29 Apple-green fluorescent cells
Adenovirus 10 VR-1087	10x LoD	34 Apple-green fluorescent cells
Adenovirus VR-14	10x LoD	37 Apple-green fluorescent cells
Adenovirus Dewitt ATCC Strain	10x LoD	15 Apple-green fluorescent cells
Adenovirus 31 VR-1109	10x LoD	42 Apple-green fluorescent cells

Clinical Performance:

Performance of the D³ FastPoint L-DFA Respiratory Virus Identification Kit testing direct respiratory specimens were established during prospective studies at 4 geographically diverse U.S. clinical laboratories during the 2009 respiratory virus seasons (January 2009 – March 2009). All specimens used in the studies meeting the inclusion and exclusion criteria represented excess, remnants of respiratory specimens that were prospectively collected from symptomatic individuals suspected of respiratory infection, and were submitted for routine care or analysis by each site, and that otherwise would have been discarded. Individual specimens were delinked from all patient identifiers and given a study sample code. All clinical sites were granted waivers of informed consent by their IRBs for this study.

Performance of the D³ FastPoint L-DFA Respiratory Virus Identification Kit was assessed and compared to a predetermined algorithm that used composite comparator methods. The composite comparator methods for influenza A virus, influenza B virus, respiratory syncytial virus, parainfluenza virus, and adenovirus consisted of Direct Specimen Fluorescent Antibody (DSFA) test with an FDA cleared device and viral culture confirmation of all the negatives (as determined by the comparator DSFA test). For human metapneumovirus the composite comparator methods consisted of DSFA with an FDA cleared device, and confirmation of all negative specimens (as determined by the comparator

DSFA test) using a validated[3] hMPV real-time RT-PCR followed by bi-directional sequencing analysis comparator assay. The hMPV real-time RT-PCR comparator assay targets the hMPV Nucleocapsid gene. "True" positive was defined as any sample that either tested positive by the comparator DSFA test or viral culture, or had bi-directional sequencing data meeting pre-defined quality acceptance criteria that matched hMPV sequences deposited in the National Center for Biotechnology Information (NCBI) GenBank database (www.ncbi.nlm.nih.gov), with acceptable E-values[4] "True" negative was defined as any sample that tested negative by both the comparator DSFA test and either viral culture or the hMPV real-time RT-PCR comparator assay.

Prevalence of the respiratory viruses within this population as determined by the D³ FastPoint L-DFA Respiratory Virus Identification Kit direct specimen testing is noted in Table 5.9 below:

Table 5.9: Respiratory Virus Prevalence

Age	Total Specimens Evaluated	Flu A	Flu B	RSV	hMPV	Adenovirus	HPIV
		# positive (prevalence)	# positive (prevalence)	# positive (prevalence)	# positive (prevalence)	# positive (prevalence)	# positive (prevalence)
0 – 1 month	55	0	0	15 (27.3%)	2 (3.6%)	0	1 (1.8%)
> 1 month to 2 years	577	27 (4.7%)	20 (3.5%)	154 (26.7%)	41 (7.1%)	11 (1.9%)	29 (5.0%)
> 2 years to 12 years	391	43 (11.0%)	104 (26.6%)	25 (6.4%)	17 (4.3%)	1 (0.3%)	6 (1.5%)
> 12 years to 21 years	173	19 (11.0%)	41 (23.7%)	4 (2.3%)	3 (1.7%)	0	2 (1.2%)
22 years to 30 years	57	3 (5.3%)	14 (24.6%)	0	1 (1.8%)	0	1 (1.8%)
31 years to 40 years	71	9 (12.7%)	9 (12.7%)	1 (1.4%)	3 (4.2%)	0	0
41 years to 50 years	52	5 (9.6%)	5 (9.6%)	0	1 (1.9%)	0	0

[3] Analytical validation of the real-time hMPV RT-PCR followed by bi-directional sequencing analysis comparator assay included analytical sensitivity and reactivity study, analytical specificity study, and extraction efficiency study. The analytical sensitivity (limit of detection or LoD) of the real-time hMPV RT-PCR followed by bi-directional sequencing analysis comparator assay was determined using quantified ($TCID_{50}$/mL) stocks of the 4 hMPV (subtypes A1, A2, B1 and B2) strains diluted in hMPV negative nasopharyngeal clinical matrix, and ranged from 10 – 50 $TCID_{50}$/mL.

[4] The E-values generated from the clinical trials range from a low of 5e-78 to a high of 1e-20. The E-Value from NCBI BLAST Alignment indicates the statistical significance of a given pair-wise alignment and reflects the size of the database and the scoring system used. The lower the E-Value, the more significant the hit. A sequence alignment that has an E-Value of 1e-3 means that this similarity has a 1 in 1000 chance of occurring by chance alone. (http://www.ncbi.nlm.nih.gov/books/bv.fcgi?rid=handbook.section.614).

Table 5.9: Respiratory Virus Prevalence							
Age	**Total Specimens Evaluated**	**Flu A**	**Flu B**	**RSV**	**hMPV**	**Adenovirus**	**HPIV**
		# positive (prevalence)	**# positive (prevalence)**	**# positive (prevalence)**	**# positive (prevalence)**	**# positive (prevalence)**	**# positive (prevalence)**
51 years to 60 years	46	3 (6.5%)	3 (6.5%)	1 (2.2%)	3 (6.5%)	0	0
61 years to 70 years	33	2 (6.1%)	2 (6.1%)	1 (3.0%)	1 (3.0%)	0	0
71 years to 80 years	16	2 (12.5%)	1 (6.3%)	1 (6.3%)	4 (25.0%)	0	0
81 years and above	7	0	0	1 (14.3%)	0	0	1 (14.3%)
Age Not Reported	41	2 (4.9%)	14 (34.1%)	0	1 (2.4%)	1 (2.4%)	0
Total	1519	115 (7.6%)	213 (14.0%)	203 (13.4%)	77 (5.1%)	13 (0.9%)	40 (2.6%)

* There were seven (7) co-infections detected: 2 - respiratory syncytial virus + metapneumovirus, 2- adenovirus + respiratory syncytial virus, 2- influenza A + metapneumovirus, 1-respiratory syncytial virus + adenovirus and 1-respiratory syncytial virus + parainfluenza virus

Tables 5.10 through 5.15 below show the study results of the NP wash/aspirate specimen type (Sites 1, 2, and 3 combined):

Table 5.10: Flu A			
Fresh nasal/nasopharyngeal wash/aspirate	Comparator DSFA (negatives followed by culture with DFA)		
DHI DSFA	Positive	Negative	Total
Positive	56	3	59
Negative	10	568	578
Total	66	571	637
			95% CI
Sensitivity	56/66	84.8%	73.9-92.5%
Specificity	568/571	99.5%	98.5-99.9%

Table 5.11: Flu B			
Fresh nasal/nasopharyngeal wash/aspirate	Comparator DSFA (negatives followed by culture with DFA)		
DHI DSFA	Positive	Negative	Total
Positive	9	0	9
Negative	2	617	619
Total	11	617	628
			95% CI
Sensitivity	9/11	81.8%	48.2-97.7%
Specificity	617/617	100.0%	99.4-100%

Table 5.12: RSV

Fresh nasal/nasopharyngeal wash/aspirate	Comparator DSFA (negatives followed by culture with DFA)		
DHI DSFA	Positive	Negative	Total
Positive	204	1	205
Negative	3	462	465
Total	207	463	670
			95% CI
Sensitivity	204/207	98.6%	95.8-99.7%
Specificity	462/463	99.8%	98.8-100%

Table 5.13: Adenovirus

Fresh nasal/nasopharyngeal wash/aspirate	Comparator DSFA (negatives followed by culture with DFA)		
DHI DSFA	Positive	Negative	Total
Positive	12	0	12
Negative	1	619	620
Total	13	619	632
			95% CI
Sensitivity	12/13	92.3%	64.0-99.8%
Specificity	619/619	100.0%	99.4-100%

Table 5.14: HPIV

Fresh nasal/nasopharyngeal wash/aspirate	Comparator DSFA (negatives followed by culture with DFA)		
DHI DSFA	Positive	Negative	Total
Positive	23	4	27
Negative	2	599	601
Total	25	603	628
			95% CI
Sensitivity	23/25	92.0%	74.0-99.0%
Specificity	599/603	99.3%	98.3-99.8%

Table 5.15: hMPV

Fresh nasal/nasopharyngeal wash/aspirate	Comparator DSFA (negatives confirmed by a validated hMPV real-time RT-PCR followed by bi-directional sequencing analysis comparator assay)		
DHI DSFA	Positive	Negative	Total
Positive	55	0	55
Negative	25	614	639
Total	80	614	694
			95% CI
Sensitivity	55/80	68.8%	57.4-78.7%
Specificity	614/614	100.0%	99.4-100%

Tables 5.16 through 5.21 below show the study results of the NP swab specimen type (Sites 3 and 4 combined):

Table 5.16: Flu A

Fresh nasal/nasopharyngeal swab	Comparator DSFA (negatives followed by culture with DFA)		
DHI DSFA	Positive	Negative	Total
Positive	57	1	58
Negative	8	623	631
Total	65	624	689
			95% CI
Sensitivity	57/65	87.7%	77.2-94.5%
Specificity	623/624	99.8%	99.1-100%

Table 5.17: Flu B

Fresh nasal/nasopharyngeal swab	Comparator DSFA (negatives followed by culture with DFA)		
DHI DSFA	Positive	Negative	Total
Positive	203	1	204
Negative	28	478	506
Total	231	479	710
			95% CI
Sensitivity	203/231	87.9%	83.7-92.1%
Specificity	478/479	99.8%	98.8-100%

Table 5.18: RSV

Fresh nasal/nasopharyngeal swab	Comparator DSFA (negatives followed by culture with DFA)		
DHI DSFA	Positive	Negative	Total
Positive	39	0	39
Negative	1	646	647
Total	40	646	686
			95% CI
Sensitivity	39/40	97.5%	86.8-99.9%
Specificity	646/646	100.0%	99.4-100%

Table 5.19: Adenovirus

Fresh nasal/nasopharyngeal swab	Comparator DSFA (negatives followed by culture with DFA)		
DHI DSFA	Positive	Negative	Total
Positive	1	0	1
Negative	0	679	679
Total	1	679	680

			95% CI
Sensitivity	1/1	100.0%	NA
Specificity	679/679	100.0%	99.5-100%

Note: The sensitivity performance of the D³ FastPoint L-DFA Respiratory Virus ID Kit detecting adenovirus from direct nasal/nasopharyngeal swab specimens has not been adequately established in the clinical study due to low adenovirus prevalence at the clinical study sites. However, the same MAb pool for adenovirus was validated in previous clinical trials for a number of FDA cleared DSFA devices. Users may wish to further evaluate the sensitivity performance of this kit detecting adenovirus using prospective nasal/nasopharyngeal swab samples.

Table 5.20: HPIV

Fresh nasal/nasopharyngeal swab	Comparator DSFA (negatives followed by culture with DFA)		
DHI DSFA	Positive	Negative	Total
Positive	13	0	13
Negative	1	667	668
Total	14	667	681
			95% CI
Sensitivity	13/14	92.9%	66.1-99.8%
Specificity	667/667	100.0%	99.4-100%

Table 5.21: hMPV

Fresh nasal/nasopharyngeal swab	Comparator DSFA (negatives confirmed by a validated hMPV real-time RT-PCR followed by bi-directional sequencing analysis comparator assay)		
DHI DSFA	Positive	Negative	Total
Positive	24	0	24
Negative	20	631	651
Total	44	631	675
			95% CI
Sensitivity	24/44	54.5%	38.8-69.9%
Specificity	631/631	100.0%	99.4-100%

Overall at the four Study Sites, the performance results of the D³ FastPoint L-DFA Respiratory Virus Identification Kit, when compared to those of the comparator devices, D³ *Ultra* Kit, D³ *Duet* RSV Kit, and D³ Metapneumovirus DFA Reagent, demonstrate that the devices detect respiratory virus antigens in a similar manner.

DEPARTMENT OF HEALTH & HUMAN SERVICES Public Health Service

Food and Drug Administration
10903 New Hampshire Avenue
Building 66
Silver Spring, MD 20993

SEP 11 2009

Mr. Ronald Lollar
Senior Director, Product Realization, Management, and Marketing
Diagnostic Hybrids Inc.
1055 East State Street Suite 100
Athens, OH 45701

Re: K091171
Trade/Device Name: D^3 FastPoint L-DFA Respiratory Virus Identification Kit
Regulation Number: 21 CFR 866.3980
Regulation Name: Respiratory viral panel multiplex nucleic acid assay
Regulatory Class: Class II
Product Code: OMG, LKT, GNX, GQS, GNY
Dated: September 2, 2009
Received: September 8, 2009

Dear Mr. Lollar:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration.

If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to such additional controls. Existing major regulations affecting your device can be found in Title 21, Code of Federal Regulations (CFR), Parts 800 to 895. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must comply with all the Act's requirements, including, but not

limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Parts 801 and 809); and good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820).

This letter will allow you to begin marketing your device as described in your Section 510(k) premarket notification. The FDA finding of substantial equivalence of your device to a legally marketed predicate device results in a classification for your device and thus, permits your device to proceed to the market.

If you desire specific information about the application of labeling requirements to your device, or questions on the promotion and advertising of your device, please contact the Office of In Vitro Diagnostic Device Evaluation and Safety at (301) 594-3084. Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21CFR Part 807.97). Other general information on your responsibilities under the Act may be obtained from the Division of Small Manufacturers, International and Consumer Assistance at its toll-free number (800) 638-2041 or (301) 443-6597 or at its Internet address http://www.fda.gov/cdrh/dsma/dsmamain.html.

Sincerely yours,

Sally A. Hojvat, M.Sc., Ph.D.
Director
Division of Microbiology Devices
Office of *In Vitro* Diagnostic Device
Evaluation and Safety
Center for Devices and
Radiological Health

Enclosure

Indications for Use

510(k) Number (if known): K091171

Device Name: D^3 FastPoint L-DFA Respiratory Virus Identification Kit

Indications For Use:

The Diagnostic Hybrids, Inc. device, D^3 FastPoint L-DFA Respiratory Virus Identification Kit is intended for the qualitative identification of influenza A virus, influenza B virus, respiratory syncytial virus, human metapneumovirus, adenovirus and to screen for the presence of parainfluenza virus types 1, 2, and 3 in nasal and nasopharyngeal swabs and aspirates/washes specimens from patients with signs and symptoms of respiratory infection by direct detection of immunofluorescence using monoclonal antibodies (MAbs).

It is recommended that specimens found to be negative for influenza A virus, influenza B virus, respiratory syncytial virus, adenovirus or parainfluenza viruses after examination of the direct specimen result be confirmed by cell culture. Specimens found to be negative for human metapneumovirus after examination of the direct specimen results should be confirmed by an FDA cleared human metapneumovirus molecular assay. Negative results do not preclude respiratory virus infection and should not be used as the sole basis for diagnosis, treatment or other management decisions.

Performance characteristics for influenza A virus detection and identification were established when influenza A (H3N2) and influenza A (H1N1) were the predominant influenza A strains circulating in the United States. Since influenza strains display antigenic drift and shift from year to year, performance characteristics may vary. If infection with a novel influenza A virus is suspected, based on clinical and epidemiological screening criteria communicated by public health authorities, collect specimens following appropriate infection control precautions and submit to state or local health departments, for testing. Viral culture should not be attempted in these cases unless a BSL 3+ facility[1] is available to receive and culture specimens.[2]

Prescription Use _X_ AND/OR Over-The-Counter Use _____
(Part 21 CFR 801 Subpart D) (21 CFR 807 Subpart C)

Division Sign-Off

Office of In Vitro Diagnostic Device Evaluation and Safety

510(k) K091171

1 www.cdc.gov

2 FDA Guidance Document: In Vitro Diagnostic Devices to Detect Influenza A Viruses: Labeling and Regulatory Path; Issued 4/10/2006

(PLEASE DO NOT WRITE BELOW THIS LINE-CONTINUE ON ANOTHER PAGE IF NEEDED)

Concurrence of CDRH, Office of In Vitro Diagnostic Devices (OIVD)