k061101

DIAGNOSTIC HYBRIDS

510 (k) Summary

November 13, 2006

NOV 20 2006

A. 510(k) Number:
k061101

B. Purpose for Submission:
New device

C. Measurand:
Influenza A, Influenza B, Respiratory Syncytial Virus (RSV), Adenovirus, Parainfluenza 1, Parainfluenza 2 and Parainfluenza 3 virus in respiratory specimens

D. Type of Test:
Direct detection or cell culture method, by immunofluorescence using fluoresceinated monoclonal antibodies (MAbs)

E. Applicant:
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F. Proprietary and Established Names:
D^3 Ultra DFA Respiratory Virus Screening & ID Kit
Common Name: DFA (Direct Fluorescent Antibody) test kit for the identification of 7 respiratory viruses (Influenza A, Influenza B, Respiratory Syncytial Virus (RSV), Adenovirus, Parainfluenza 1, Parainfluenza 2 and Parainfluenza 3 virus) in patient specimens and cell cultures

G. Regulatory Information:
1. Regulation section:
   866.3330 Influenza virus serological reagents
2. Classification:
   Class I
3. Product code:
   GNW
4. Panel:
   Microbiology (83)

H. Intended Use:
1. Intended use(s):
   The Diagnostic Hybrids, Inc. D3 Ultra DFA (direct fluorescent antibody) RESPIRATORY VIRUS 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 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.

2. Indication(s) for use:
   The Diagnostic Hybrids, Inc. D3 Ultra DFA (direct fluorescent antibody) RESPIRATORY VIRUS 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 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.
3. Special conditions for use statement(s):
   For prescription use only
4. Special instrument requirements:
   Fluorescence microscope with the correct filter combination for FITC (excitation peak = 490 nm, emission peak = 520nm).

I. Device Description:

The Diagnostic Hybrids, Inc. D3 *Ultra* DFA RESPIRATORY VIRUS SCREENING & ID KIT includes a DFA Screening Reagent that contains a blend of murine monoclonal antibodies (MAbs) directed against seven respiratory viruses (Influenza A, Influenza B, Respiratory Syncytial Virus, Adenovirus, Parainfluenza 1, Parainfluenza 2, and Parainfluenza 3) plus seven separate DFA Reagents, each consisting of MAb blends directed against a single respiratory virus. The kit can be used for direct specimen or cell culture screening and final virus identification.

Kit Components:

- Respiratory Virus DFA Screening Reagent - a mixture of fluorescein labeled murine monoclonal antibodies directed against respiratory viral antigens of Influenza A, Influenza B, Respiratory Syncytial Virus (RSV), Adenovirus, Parainfluenza 1, Parainfluenza 2 and Parainfluenza 3. The buffered, stabilized, aqueous solution contains Evan's Blue as a counter-stain and 0.1% sodium azide as preservative.
- Influenza A DFA Reagent - fluorescein labeled murine monoclonal antibodies directed against antigens produced by Influenza A virus infected cells. The buffered, stabilized, aqueous solution contains Evan's Blue as a counter-stain and 0.1% sodium azide as preservative.
- Influenza B DFA Reagent - fluorescein labeled murine monoclonal antibodies directed against antigens produced by Influenza B virus infected cells. The buffered, stabilized, aqueous solution contains Evan's Blue as a counter-stain and 0.1% sodium azide as

preservative.

- RSV DFA Reagent - fluorescein labeled murine monoclonal antibodies directed against antigens produced by RSV infected cells. The buffered, stabilized, aqueous solution contains Evan's Blue as a counter-stain and 0.1% sodium azide as preservative.
- Adenovirus DFA Reagent - fluorescein labeled murine monoclonal antibodies directed against antigens produced by Adenovirus infected cells. The buffered, stabilized, aqueous solution contains Evan's Blue as a counter-stain and 0.1% sodium azide as preservative.
- Parainfluenza 1 DFA Reagent - fluorescein labeled murine monoclonal antibodies directed against antigens produced by Parainfluenza 1 infected cells. The buffered, stabilized, aqueous solution contains Evan's Blue as a counter-stain and 0.1% sodium azide as preservative.
- Parainfluenza 2 DFA Reagent - fluorescein labeled murine monoclonal antibodies directed against antigens produced by Parainfluenza 2 infected cells. The buffered, stabilized, aqueous solution contains Evan's Blue as a counter-stain and 0.1% sodium azide as preservative.
- Parainfluenza 3 DFA Reagent - fluorescein labeled murine monoclonal antibodies directed against antigens produced by Parainfluenza 3 infected cells. The buffered, stabilized, aqueous solution contains Evan's Blue as a counter-stain and 0.1% sodium azide as preservative.
- Respiratory Virus Antigen Control Slides - five individually packaged control slides containing 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, Influenza B, Respiratory Syncytial Virus (RSV), Adenovirus, Parainfluenza 1, Parainfluenza 2 and Parainfluenza 3. The Negative well contains uninfected cells. Each slide is intended to be stained only one time.
- Normal Mouse Gamma Globulin DFA Reagent - a mixture of fluorescein labeled murine gamma globulin that has been shown to be unreactive with any of the listed respiratory viruses. The buffered, stabilized, aqueous solution contains Evan's Blue as a counter-stain and 0.1% sodium azide as preservative.
- Wash Solution Concentrate - a 40X concentrate consisting of Tween 20 and 4% sodium azide (after dilution to 1X in water, the concentration of sodium azide in the solution is 0.1%) in Phosphate Buffered Saline.
- Mounting Fluid - an aqueous, buffered, stabilized solution of glycerol and 0.1% sodium azide.

J. Substantial Equivalence Information:

1. Predicate device name(s):
   DFA Respiratory Virus Screening & ID Kit
2. Predicate 510(k) number(s):
   K022713
3. Comparison with predicate:
   The similarities to predicate device are in indicated use, operating principle, basic design, materials and formulation.

Similarities		
Item	Device	Predicate
Intended Use	For the qualitative detection and identification of the respiratory viruses, Influenza A, Influenza B, Respiratory Syncytial Virus (RSV), Adenovirus, Parainfluenza 1, Parainfluenza 2 and Parainfluenza 3 virus by either direct detection or cell culture method	For the qualitative detection and identification of the respiratory viruses, 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
Basic principle	DFA (Direct Fluorescent Antibody) test - Immunofluorescence using fluoresceinated monoclonal antibodies (MAbs)	DFA (Direct Fluorescent Antibody) test - Immunofluorescence using fluoresceinated monoclonal antibodies (MAbs)
Antibody	Blend of murine monoclonal antibodies (MAbs) directed against seven respiratory viruses	Blend of murine monoclonal antibodies (MAbs) directed against seven respiratory viruses
Instrumentation (required but not provided)	Fluorescence microscope with the correct filter combination for FITC (excitation peak = 490 nm, emission peak = 520nm).	Fluorescence microscope with the correct filter combination for FITC (excitation peak = 490 nm, emission peak = 520nm).
Sample type	Respiratory specimens	Respiratory specimens

K. Standard/Guidance Document Referenced (if applicable):

N/A

L. Test Principle:

The test kit uses viral antigen-specific murine monoclonal antibodies that are directly labeled with fluorescein for rapid detection and identification of respiratory viruses. The cells to be tested, either derived from a clinical specimen or cell culture, are fixed in acetone. The DFA Screening Reagent is added to the cells to determine the presence of viral antigens. After incubating at 35°C to 37°C, the stained cells are rinsed with the diluted Wash Solution, a drop of the supplied Mounting Fluid is added and a coverslip is placed on the prepared cells. The cells are examined using a fluorescence microscope. Virus infected cells will be stained with viral specific apple-green fluorescence when stained with the DFA

Screening Reagent while uninfected cells will contain no fluorescence but will be stained red by the Evan's Blue counter-stain. If on examination of a *direct stained* specimen, no fluorescent-stained cells are found and all the cells stain red from the Evan's Blue, it is recommended that the specimen be cultured and stained using the DFA Screening Reagent. If fluorescent cells are seen, the particular virus is identified using the separate DFA Reagents on new, separate cell preparations. Cell preparations are fixed in acetone. The individual virus DFA Reagents are added to the cell preparations. After incubating at 35° to 37°C, the stained cells are rinsed with the diluted Wash Solution, a drop of the supplied Mounting Fluid is added and a coverslip is placed on the stained cells. The cells are examined using a fluorescence microscope for the presence of viral specific apple-green fluorescence, by which the unknown respiratory virus is identified.

Interpretation of results:

It is recommended that controls be examined first to ensure proper test performance before examination of the specimens. A positive reaction is one in which bright apple-green fluorescence is observed in the infected cells. Uninfected cells will stain dull red due to the Evan's Blue counter-stain included in the DFA Reagent.

Fluorescent staining pattern of respiratory virus infected cells:

The "typical" apple-green fluorescence staining pattern for each virus is as follows:

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 cytoplasmic and punctate with small inclusions in the syncytia.

Adenovirus: The fluorescence is cytoplasmic and punctate or bright nuclear or both.

Parainfluenza 1, 2, 3: The fluorescence is cytoplasmic and punctate with irregular inclusions. Types 2 and 3 cause the formation of syncytia.

Co-infection with more than one infecting virus present in the specimen has been reported in a number of studies. The presence of multiple viruses is indicated when more than one well of the 8-well slide has fluorescent cells. The identification of the viruses is based on the individual virus DFA Reagents showing fluorescence. In such a case, it should be reported as "... and ... detected by direct specimen testing." or "... and ... isolated by cell culture."

Results From Direct Specimen Testing: The quality of the specimen with respect to the number of epithelial cells in the sample can be assessed by examining the different fields at a magnification of 200X. A satisfactory specimen should have at least 2 columnar epithelial cells per field. A negative result is indicated by the absence of fluorescence in a minimal sampling of 20 columnar epithelial cells. An inadequate sample is indicated by fewer than 20 columnar epithelial cells present in the sample, in which case the test is considered invalid. A new specimen should be obtained and tested or cell culture of the remaining specimen should be initiated.

A satisfactory specimen with no fluorescent cells found should be reported as "Presumptively negative, no Influenza A, Influenza B, Adenovirus, Respiratory Syncytial Virus, Parainfluenza 1, Parainfluenza 2, or Parainfluenza 3 detected by direct specimen testing". However, such negative results should be confirmed using cell culture. In the case culture yields a positive result, it should be reported as "... isolated by cell culture", where '...' is the appropriate virus.

If fluorescent cells are found, continue with the Testing Procedure, staining with the individual virus DFA Reagents. The individual virus DFA Reagent that yields fluorescent cells represents the identification of the respiratory virus. In such a case, it should be reported as "... detected by direct specimen testing", where '...' is the appropriate virus, e.g. Influenza A, Influenza B, Adenovirus, Respiratory Syncytial Virus, Parainfluenza 1, Parainfluenza 2, or Parainfluenza 3.

Results from Culture Isolation / Confirmation: The entire cell spot or monolayer of cells must be examined for virus-infected, fluorescent cells. If no fluorescent cells are found, the results of testing of the specimen should be reported as, "No Influenza A, Influenza B, Adenovirus, Respiratory Syncytial Virus, Parainfluenza 1, Parainfluenza 2, or Parainfluenza 3 isolated by cell culture."

If fluorescent cells are found, continue with the Testing Procedure, staining with the individual virus DFA. The individual virus DFA Reagent that yields fluorescent cells represents the identification of the respiratory virus. In such a case, it should be reported as "... isolated by cell culture", where '...' is the appropriate virus, e.g. Influenza A, Influenza B, Respiratory Syncytial Virus, Parainfluenza 1, Parainfluenza 2, Parainfluenza 3, or Adenovirus.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:
   a. *Precision/Reproducibility:*
      Not applicable
   b. *Linearity/assay reportable range:*
      Not applicable
   c. *Traceability, Stability, Expected values (controls, calibrators, or methods):*
      Not applicable
   d. *Detection limit:*
      The Predicate and Subject MAbs were compared by inoculating 96-well cell culture plates with the appropriate virus stock diluted to give ~ 50 viral particles for the entire plate or 1 viral particle per every 2 wells (i.e. 1 TCID50). The plates were incubated at 37° C for 24 hours and then stained with either the Subject Kit or the Predicate Kit. All plates were stained using the procedure in the product insert. This assay was performed 3 times with each virus and the results indicate no statistical difference from the Predicate kit by a paired t-test.

      Each targeted respiratory virus was diluted to a value of 250 virus/mL and serial 2-fold dilutions were then done down to a final value of 0.49 virus/mL. Each dilution of virus was inoculated into 6 shell vials of R-Mix, centrifuged at 700xg for 60 minutes and incubated at 35-37°C for 48 hours. The Subject Kit or the Predicate Kit was used to stain 3 shell vials of each viral dilution according to the product insert. The sensitivity of both fluorescent antibody stains is equivalent, with ~ 0.5 – 1.0 PFU as the minimum viral dilution detected.

      In order to determine if mixing the 15 various MAbs in the Screening Reagent affected their sensitivity, the analytical sensitivity of the subject screen reagent was compared to that of the subject individual reagents by inoculating 96-well cell culture plates with the appropriate virus stock diluted to give ~ 50 viral particles for

the entire plate or 1 viral particle per every 2 wells. The plates were incubated at 37°C for 24 hours and then stained with either the subject screening reagent or the subject individual reagents. All plates were stained using the procedure in the product insert. This assay was performed 3 times with each virus and the results indicate that the sensitivity of the screening reagent in the subject kit was not statistically different from that of the subject individual reagents using the paired test.

Analytical specificity:

The DFA Reagents were tested for cross-reactivity against a wide variety of cells and microorganisms. No cross-reactivity was observed for 64 virus strains (cultured and processed for staining) or for 18 host culture cell types. Eighteen (18) bacterial cultures were stained and examined for cross-reactivity, including *Staphylococcus aureus*, a protein-A-producing bacterium. Staining of *S. aureus* appeared as small points of fluorescence while all other bacterial cultures were negative. [Protein A will nonspecifically bind to the Fc portions of conjugated antibodies. Such binding can be distinguished from viral antigen binding on the basis of morphology, i.e., *S. aureus*-bound fluorescence appears as small (~1 micron diameter), bright dots. Stringent conditions for cross-reactivity testing were achieved by using high concentration DFAs and high titers of microorganisms. The DFAs (i.e. directly fluoresceinated monoclonal antibodies) were prepared at 1.5X the concentration that is provided in the kit. Each of the tested viruses was prepared as infected cell monolayers (250 infected cells inoculated into a shell vial culture and incubated for 24 to 48 hours, to yield a 3+ to 4+ infection), and processed and stained with the 1.5X DFAs according to the procedure detailed in the product insert. Bacterial strains were cultured, processed as suspensions, then spotted on microscope slides (yielding > 150 bacteria per 400X microscope field), then stained with the 1.5X DFAs according to the procedure in the product insert. Cell cultures were stained as confluent monolayers.

		DFA Reagent (Results Positive (+) or Negative (-) for Reactivity)						
Organism	Strain	Adeno	Flu A	Flu B	Para 1	Para 2	Para 3	RSV
Adenovirus	Type 1	+	-	-	-	-	-	-
	Type 3	+	-	-	-	-	-	-
	Type 5	+	-	-	-	-	-	-
	Type 6	+	-	-	-	-	-	-
	Type 7	+	-	-	-	-	-	-
	Type 10	+	-	-	-	-	-	-
	Type 13	+	-	-	-	-	-	-
	Type 14	+	-	-	-	-	-	-
	Type 18	+	-	-	-	-	-	-
	Type 31	+	-	-	-	-	-	-
	Type 40	+	-	-	-	-	-	-
	Type 41	+	-	-	-	-	-	-
Influenza A	Aichi (H3N2)	-	+	-	-	-	-	-
	Mal (H1N1)	-	+	-	-	-	-	-
	Hong Kong (H3N2)	-	+	-	-	-	-	-
	Denver (H1N1)	-	+	-	-	-	-	-
	Port Chalmers (H3N2)	-	+	-	-	-	-	-
	Victoria (H3N2)	-	+	-	-	-	-	-
	New Jersey ($H_{sw}N1$)	-	+	-	-	-	-	-
	WS (H1N1)	-	+	-	-	-	-	-

	PR (H1N1)	-	+	-	-	-	-	-
Influenza B	Hong Kong	-	-	+	-	-	-	-
	Maryland	-	-	+	-	-	-	-
	Mass	-	-	+	-	-	-	-
	Taiwan	-	-	+	-	-	-	-
	GL	-	-	+	-	-	-	-
	Russia	-	-	+	-	-	-	-
RSV	Long	-	-	-	-	-	-	+
	Wash	-	-	-	-	-	-	+
	9320	-	-	-	-	-	-	+
Rhinovirus 39	209 Picornavirus	-	-	-	-	-	-	-
Parainfluenza 1	C-35	-	-	-	+	-	-	-
Parainfluenza 2	Greer	-	-	-	-	+	-	-
Parainfluenza 3	C 243	-	-	-	-	-	+	-
Parainfluenza 4a	M-25	-	-	-	-	-	-	-
Parainfluenza 4b	CH19503	-	-	-	-	-	-	-
Metapneumovirus	A1	-	-	-	-	-	-	-
	A2	-	-	-	-	-	-	-
	B3	-	-	-	-	-	-	-
	B4	-	-	-	-	-	-	-
Coronavirus	OC43	-	-	-	-	-	-	-
	229E	-	-	-	-	-	-	-
Herpes simplex virus Type 1	1F	-	-	-	-	-	-	-
	MacIntyre	-	-	-	-	-	-	-
Herpes simplex virus Type 2	MS	-	-	-	-	-	-	-
	Strain G	-	-	-	-	-	-	-
Cytomegalovirus	Towne	-	-	-	-	-	-	-
	Davis	-	-	-	-	-	-	-
	AD169	-	-	-	-	-	-	-
Varicella-zoster	Webster	-	-	-	-	-	-	-
	Ellen	-	-	-	-	-	-	-
Echovirus	9	-	-	-	-	-	-	-
	11	-	-	-	-	-	-	-
	30	-	-	-	-	-	-	-
	34	-	-	-	-	-	-	-
Coxsackievirus	B1	-	-	-	-	-	-	-
	B2	-	-	-	-	-	-	-
	B3	-	-	-	-	-	-	-
	B4	-	-	-	-	-	-	-
	B5	-	-	-	-	-	-	-
	B6	-	-	-	-	-	-	-
Mumps		-	-	-	-	-	-	-
Rubeola		-	-	-	-	-	-	-
Rhinovirus	209 Picornavirus	-	-	-	-	-	-	-
Achoieplasma laidlawii		-	-	-	-	-	-	-
Bordetella bronchiseptica		-	-	-	-	-	-	-
Bordetella pertussis		-	-	-	-	-	-	-
Chlamydia pneumoniae		-	-	-	-	-	-	-
Clostridium diphtheriae		-	-	-	-	-	-	-
Haemophillus influenzae type A		-	-	-	-	-	-	-
Klebsiella pneumoniae		-	-	-	-	-	-	-
Listeria pneumophila		-	-	-	-	-	-	-
Moraxella cartarrhalis		-	-	-	-	-	-	-
Mycobacterium avium		-	-	-	-	-	-	-
Mycobacterium intracellulare		-	-	-	-	-	-	-
Mycoplasma hominis type 1		-	-	-	-	-	-	-
Mycoplasma orale		-	-	-	-	-	-	-
Mycoplasma pneumoniae		-	-	-	-	-	-	-
Mycoplasma salivarium		-	-	-	-	-	-	-
Pseudomonas aeruginosa		-	-	-	-	-	-	-
Streptococcus pneumoniae		-	-	-	-	-	-	-

Streptococcus pyogenes	-	-	-	-	-	-	-
Ureaplasma urealyticum	-	-	-	-	-	-	-
Cell cultures:							
A549	-	-	-	-	-	-	-
BGMK	-	-	-	-	-	-	-
HEp-2	-	-	-	-	-	-	-
LLC-MK2	-	-	-	-	-	-	-
MDCK	-	-	-	-	-	-	-
MRC-5	-	-	-	-	-	-	-
MRHF	-	-	-	-	-	-	-
Mv1Lu	-	-	-	-	-	-	-
NCI-H292	-	-	-	-	-	-	-
pCMK	-	-	-	-	-	-	-
pRhMK	-	-	-	-	-	-	-
pRK	-	-	-	-	-	-	-
RD	-	-	-	-	-	-	-
RhMK II	-	-	-	-	-	-	-
R-Mix™	-	-	-	-	-	-	-
R-Mix™ Too	-	-	-	-	-	-	-
Vero	-	-	-	-	-	-	-
WI-38	-	-	-	-	-	-	-

f. Assay cut-off:

Not applicable

2. Comparison studies:

a. Method comparison with predicate device:

The study included 849 original specimens evaluated by this product ("Subject" test) and a currently marketed DFA Screening & Identification Kit ("Predicate" test). All 849 specimens were studied by Direct Specimen (DS) testing with 22 of these specimens having insufficient cell numbers to be evaluated, and one other which could not be evaluated because it exhibited non-specific staining from the Normal Mouse Gamma Globulin DFA Reagent; 520 of the specimens also were studied by Cell Culture (CC) method with one specimen not evaluated because it produced a toxic cell culture monolayer. All but 30 of the specimens were prospectively collected during the 2005-2006 season; those 30 specimens had been archived as Parainfluenza-positive. In addition, a set of 81 clinical isolates were tested by CC methods only. The evaluations were conducted at three laboratory sites. Summary of results are shown in tables below:

Direct specimen testing – fresh prospectively collected specimens:									
326 specimens	Negative	Screen +	Adeno virus	Influenza A	Influenza B	Parainfluenza 1	Parainfluenza 2	Parainfluenza 3	Respiratory Syncytial Virus
Predicate Results:	236	90	18	32	18	2	0	2	18
Subject Results:	232	94	18	32	19	2	0	5	18
Positive Percent Agreement (PPA)		95.5%	100.0%	100.0%	100.0%	100.0%	---	100.0%	100.0%
95% Confidence Interval – PPA		89.0-98.2%	82.4-100%	89.3-100%	82.4-100%	34.2-100%	---	34.2-100%	82.4-100%
Negative Percent Agreement (NPA)	98.3%		100.0%	100.0%	98.7%	100.0%	100.0%	96.7%	100.0%
95% Confidence Interval - NPA	95.7-99.3%		95.2-100%	94.2-100%	92.9-99.8%	96.0-100%	96.1-100%	90.8-98.9%	95.2-100%

Direct specimen testing – frozen prospectively collected specimens:									
474 specimens	Negative	Screen +	Adeno virus	Influenza A	Influenza B	Parainfluenza 1	Parainfluenza 2	Parainfluenza 3	Respiratory Syncytial Virus
Predicate Results:	306	168	8	85	19	3	3	9	51
Subject Results:	306	168	8	85	19	3	3	9	51
PPA		100%	100%	100%	100%	100%	100%	100%	100%
95% CI – PPA		97.8-100%	63.1-100%	95.7-100%	82.3-100%	38.3-100%	38.3-100%	70.1-100%	93.0-100%
NPA	100%		100%	100%	100%	100%	100%	100%	100%
95% CI – NPA	98.8-100%		97.7-100%	95.6-100%	97.6-100%	97.8-100%	97.8-100%	97.6-100%	96.7-100%

Cell culture testing – frozen prospectively collected specimens:									
490 specimens	Negative	Screen +	Adeno virus	Influenza A	Influenza B	Parainfluenza 1	Parainfluenza 2	Parainfluenza 3	Respiratory Syncytial Virus
Predicate Results:	309	181	13	93	23	6	4	9	49
Subject Results:	309	181	13	93	23	6	4	9	49
PPA		100%	100%	100%	100%	100%	100%	100%	100%
95% CI – PPA		98.0-100%	73.4-100%	95.2-100%	83.1-100%	55.7-100%	45.4-100%	65.5-100%	91.3-100%
NPA	100%		100%	100%	100%	100%	100%	100%	100%
95% CI – NPA	98.5 – 100%		97.3-100%	95.0-100%	97.1-100%	97.4-100%	97.4-100%	96.6-100%	96.6-100%

Specimens and culture isolates used in these studies came from nasopharyngeal (NP) aspirates, washes, swabs, bronchial alveolar lavages (BAL) and/or tracheal aspirates.

b. Matrix comparison:
n/a

3. Clinical studies:
   a. Clinical Sensitivity:
   Not applicable.
   b. Clinical specificity:
   Not applicable.
   c. Other clinical supportive data (when a. and b. are not applicable):

Non-prospective archival specimens: Due to relative low prevalence of Parainfluenza infections in populations of respiratory specimens, few prospectively collected specimens were reactive with the Parainfluenza DFA Reagents. Frozen original specimens previously determined to contain Parainfluenza (types 1, 2, or 3) during the 2006 "respiratory season" were obtained from an additional laboratory, and were tested in an internal reference laboratory using the Subject and Predicate Tests by Direct Specimen method (Study 3a-DS; see table below). The same specimens were tested by Cell Culture method (Study 3a-CC, see table below). Original results reported by the laboratory were unknown to the study investigator. Although the study design has a

selection bias, this study offers further analytical information on the assay's ability to detect Parainfluenza viruses.

Study 3a-DS – Direct Specimen Results									
26 specimens	Negative	Screen +	Adeno virus	Influenza A	Influenza B	Parainfluenza 1	Parainfluenza 2	Parainfluenza 3	Respiratory Syncytial Virus
Predicate Results	9	17	0	0	0	1	5	11	0
Subject Results:	8	18	0	0	0	1	5	12	0
PPA		100%	---	---	---	100%	100%	100%	---
95% CI – PPA		78.4% - 100%	---	---	---	16.8% - 100%	51.1% - 100%	70.0% - 100%	---
NPA	88.9%		100%	100%	100%	100%	100%	85.7%	100%
95% CI – NPA	54.3% - >99.9%		79.3% - 100%	79.3% - 100%	79.3% - 100%	78.4% - 100%	73.4% - 100	46.7% - 99.5%	79.3% - 100%

Study 3a-CC – Cell Culture Results									
29 specimens	Negative	Screen +	Adeno virus	Influenza A	Influenza B	Parainfluenza 1	Parainfluenza 2	Parainfluenza 3	Respiratory Syncytial Virus
Predicate Results	8	21	0	0	0	3	5	13	0
Subject Results:	8	21	0	0	0	3	5	13	0
PPA		100%	---	---	---	100%	100%	100%	---
95% CI – PPA		81.8% - 100%	---	---	---	38.3% - 100%	51.1% - 100%	73.4% - 100%	---
NPA	100%		100%	100%	100%	100%	100%	100%	100%
95% CI – NPA	62.8% - 100%		81.8% - 100%	81.8% - 100%	81.8% - 100%	79.3% - 100%	77.3% - 100	62.8% - 100%	81.8% - 100%

Non-prospective archival clinical isolates: A study was conducted using a collection of banked clinical isolates known to contain respiratory viruses that had been frozen from the 2005/2006 respiratory season. These specimens were selected because they were previously shown to contain at least one of the seven virus analytes detected by the Subject Test.

Study 3b-CC – Cell Culture Results									
81 specimens	Negative	Screen +	Adeno virus	Influenza A	Influenza B	Parainfluenza 1	Parainfluenza 2	Parainfluenza 3	Respiratory Syncytial Virus
Predicate Results:	0	81	11	18	17	4	1	26	5
Subject Results:	0	81	11	18	17	4	1	26	5
PPA		100%	100%	100%	100%	100%	100%	100%	100%
95% CI – PPA		94.6% - 100%	70.0% - 100%	79.3% - 100%	78.4% - 100%	45.4% - 100%	16.8% - 100%	84.8% - 100%	51.1% - 100%
NPA		100%	100%	100%	100%	100%	100%	100%	100%
95% CI – NPA		97.3% - 100%	93.8% - 100%	93.1% - 100%	93.2% - 100%	94.3% - 100%	94.5% - 100	92.2% - 100%	94.2% - 100%

4. Clinical cut-off:
   Not applicable
5. Expected values/Reference range:
   Respiratory virus infections are often seasonal, with Influenza typically extending from

November to April in the northern hemisphere, and Adenovirus infections occurring more often during late winter to early summer. RSV is usually a seasonal (winter and early spring) infection as well, with epidemics lasting up to 5 months, while outbreaks caused by parainfluenza viruses may occur throughout a year.
The clinical studies were comprised of respiratory specimens collected during the winter to early spring months of 2005/2006. Prevalence of the respiratory viruses within the population of specimens that were prospectively collected and tested fresh is noted in the table below:

Expected Values	Adenovirus	Influenza A	Influenza B	Parainfluenza 1	Parainfluenza 2	Parainfluenza 3	Respiratory Syncytial Virus
Fresh Specimens (n = 326)	18	32	19	2	0	5	18
Prevalence	5.5%	9.8%	5.8%	0.6%	0	1.5%	5.5%

The following table summarizes the participant age demographics according to test results for a population of 326 fresh specimens, prospectively collected and evaluated for performance using the predicate assay:

Virus: / Age❖:	Adenovirus	Influenza A	Influenza B	Parainfluenza 1	Parainfluenza 2	Parainfluenza 3	Respiratory Syncytial Virus	Negative
Totals	**18**	**32**	**18**	**2**	**0**	**2**	**18**	**236**
<1m	0	0	0	0	0	0	2	1
1m to 2y	8	9	4	1	0	2	8	80
2y to12y	8	7	6	0	0	0	1	42
12y to18y	1	1	5	0	0	0	0	8
18y to 21y	0	0	1	0	0	0	0	2
>21y	0	12	1	0	0	0	1	78
Not reported	1	3	1	1	0	0	6	25

❖ *Age: m = months, and y = years*

DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service

Food and Drug Administration
2098 Gaither Road
Rockville MD 20850

NOV 20 2006

Ms. Gail R. Goodrum
Vice President, Regulatory and Quality Affairs
Diagnostic Hybrids, Inc.
350 West State Street
Athens, OH 45701

Re: k061101
Trade/Device Name: D^3 Ultra DFA Respiratory Virus Screening & ID Kit
Regulation Number: 21 CFR 866.3330
Regulation Name: Influenza virus serological reagents
Regulatory Class: Class I
Product Code: GNW
Dated: October 10, 2006
Received: October 11, 2006

Dear Ms. Goodrum:

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.

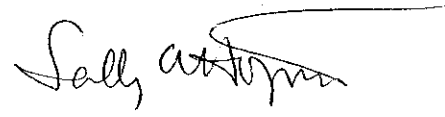
If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to such additional controls. Existing major regulations affecting your device can be found in Title 21, Code of Federal Regulations (CFR), Parts 800 to 895. In addition, FDA may publish 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 (240)276-0450. 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):_____k061101

Device Name:_____D^3 Ultra DFA Respiratory Virus Screening & ID Kit

Indications For Use:

The Diagnostic Hybrids, Inc.'s D^3 *Ultra* DFA (direct fluorescent antibody) RESPIRATORY VIRUS 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 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.

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

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

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

Division Sign-Off

Office of In Vitro Diagnostic Device Evaluation and Safety

510(k) k061101