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# **Certificate of Analysis**

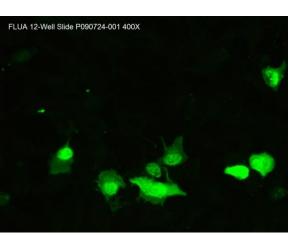
# **INFLUENZA VIRUS TYPE A**

FA Substrate Slide

CATALOG NO.: SLD-IFA-FLUA SIZE: 12 Well LOT: P090724-001 EXPIRATION: 31 March 2025

**AGENT:** Influenza Virus Type A **ISOLATE:** ABR Equine

CELL CULTURE SUBSTRATE: Mink lung cells



**DESCRIPTION:** Slides are virus-infected cell cultures grown on the surface of Teflon-masked slides. They are supplied fixed and unstained in moisture-free foil pouches. All wells contain both positive and negative cells. The positive cells usually total no more than 30% of the cells in the well so that good contrast may be seen.

**QUALITY CONTROL METHOD:** Indirect FA using an equine (in house sample) for a positive control and Anti-Equine IgG FITC (catalog no. CJ-F-EQUG-1ML or 10ML).

**Specific Reaction:** 3-4+ signal with the positive serum sample at 1/50, no background.

Other Comments: 10-30 infected cells per high powered field.

**PATTERN OF FLUORESCENCE:** Individual cells with pronounced plasma membrane fluorescence and some fluorescing cytoplasmic and nuclear inclusion bodies.

**INTENDED USE:** Generally used for Indirect FA to detect antibody to Influenza Virus Type A but may also be used as a control substrate slide for Direct FA conjugates when applicable.

**STORAGE:** Store sealed in foil pouch at -20°C. Avoid self-defrosting freezers.

**REFERENCES:** NA

## FOR IN VITRO LABORATORY USE ONLY.

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H:\Master Documents\COA (VMRD)\8-, 10-, 12-well Slides\FLUA\SLD-IFA-FLUA\_P090724-001\_250331.docx 12 April 2021

#### **RECOMMENDED STAINING PROCEDURE FOR INDIRECT FA:**

- 1. Warm slide to room temperature before removing from foil pouch.
- 2. Place diluted serum on the designated wells. Dilute serum in serum diluting buffer, pH 7.2 (catalog no. SSDB-100ML).
- 3. Incubate slide in humid chamber at 37°C for 30 minutes.
- 4. Using a wash bottle, gently rinse slide briefly in FA rinse buffer, pH 9.0 (catalog no. FARB-4X) and then soak for 10 minutes in FA rinse buffer, pH 9.0.
- 5. Drain slide and dry around wells by pressing blotter (included in pouch) to front surface. Place labeled anti-IgG or IgM on the wells.
- 6. Incubate as in step 3.
- 7. Rinse as in step 4.
- 8. Drain slide and dry <u>back and edges</u> with a paper towel. Do not allow stained surface to dry. Do not rinse with water.
- 9. Mount with mounting fluid [glycerol/FA rinse buffer, pH 9.0, (50/50)] (catalog no. FAMF-10ML) and view with good quality fluorescence microscope at 100X-250X. Confirmation may be made at 400X.

#### **RECOMMENDED STAINING PROCEDURE FOR DIRECT FA:**

- 1. Warm slide to room temperature before removing from foil pouch.
- 2. Place direct FA conjugate on the designated wells.
- 3. Incubate slide in humid chamber at 37°C for 30 minutes.
- 4. Using a wash bottle, gently rinse slide briefly in FA rinse buffer, pH 9.0 (catalog no. FARB-4X) and then soak for 10 minutes in FA rinse buffer, pH 9.0.
- 5. Drain slide and dry <u>back and edges</u> with a paper towel. Do not allow stained surface to dry. Do not rinse with water.
- 6. Mount with mounting fluid [glycerol/FA rinse buffer, pH 9.0, (50/50)] (catalog no. FAMF-10ML) and view with good quality fluorescence microscope at 100X-250X. Confirmation may be at 400X.

## SERUM DILUTING BUFFER (pH 7.2):\*

-	Na <sub>2</sub> HPO <sub>4</sub> 1.19	gm
-	NaH <sub>2</sub> PO <sub>4</sub> 0.22	gm
	NaCl	
-	BSA	gm
	$DI/dH_2O\ldots\ldots Q.S.$	

\* This recipe makes 1 liter. If you need less, adjust recipe accordingly. Store at 2-7°C. Add 0.09% NaN<sub>3</sub> if diluted serum is not going to be used within one week.

#### 4X FA RINSE BUFFER (pH 9.0):

- Na <sub>2</sub> CO <sub>3</sub>	11.4 gm
- NaHCO <sub>3</sub>	
- NaCl	
- DI/dH <sub>2</sub> O	Q.S. to 1 liter

Final pH should be 9.0-9.5. This is a 4X concentrate and should be diluted 1/4 with DI/distilled water for use as a working buffer. Keep in a tightly stoppered container at room temperature. MOUNTING FLUID is made by mixing glycerol and FA rinse buffer, pH 9.0, in equal proportions.