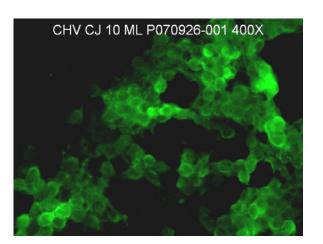


Certificate of Analysis

Canine Herpesvirus (CHV) Direct FA Conjugate

CATALOG NO.: 210-13-CHV VOLUME: 10 ml LOT: P070926-001 EXPIRATION: 31 July 2011

VIRUS: Canine Herpesvirus (CHV)



DESCRIPTION: Anti-CHV polyclonal antiserum conjugated to fluorescein isothiocyanate. Canine origin. Ready to use.

QUALITY CONTROL METHOD: Direct FA on CHV-infected cell cultures using VMRD, Inc. FA substrate slides (catalog no. SLD-IFA-CHV) to detect binding.

Specific Reaction:3-4+ on positive cells with no background.Other Reactions or Comments:NA

INTENDED USE: This reagent is suitable for CHV virus identification in cell cultures and in animal tissues. Reacts with Canine Parainfluenza Virus (CPI). Definitive diagnosis can be made using CPI Conjugate (catalog no. 210-33-CPI2) which does not cross-react with Canine Herpesvirus.

STORAGE: This conjugate is provided in liquid form and should be stored at 4-8°C. DO NOT FREEZE! If conjugate becomes cloudy, it should be discarded. This conjugate contains 10 ppm ProClin 300 as a preservative.

FOR IN VITRO LABORATORY USE ONLY.

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RECOMMENDED STAINING PROCEDURE FOR DIRECT FA:

- 1. Air dry smears or tissue sections for at least 30 minutes at room temperature (do not dry cell cultures!).
- 2. Fix smears or tissue sections on slides for 20 minutes in acetone-methanol (75/25) at room temperature. Cell cultures should be rinsed with PBS and fixed in pure acetone for 10 minutes at room temperature. After fixation and before staining, slides should be dried for 10 minutes in a dry 37°C incubator.
- 3. Stain slides with 50-75 µl conjugate for 30 minutes at 37°C in humid chamber.
- 4. Gently rinse slides briefly in FA Rinse Buffer, pH 9.0 (VMRD catalog no. 210-90-RB) and then soak for 10 minutes in FA Rinse Buffer, pH 9.0.
- 5. Drain slides 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 FA Mounting Fluid [glycerol/FA rinse buffer, pH 9.0, (50/50)] (VMRD catalog no. 210-92-MF) and view with good quality fluorescence microscope at 100X-250X. Confirmation may be made at 400X.

PHOSPHATE BUFFERED SALINE (PBS) SOLUTION (pH 7.2):

- Na ₂ HPO ₄	.19 gm
- NaH ₂ PO ₄ 0	.22 gm
- NaCl	5.55 gm
- DI/dH ₂ O	o 1 liter

4X FA RINSE BUFFER (pH 9.0):

- Na ₂ CO ₃	11.4 gm
- NaHCO ₃	
- NaCl	8.5 gm
- DI/dH ₂ 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.