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## Certificate of Analysis

### **Canine Coronavirus (CCV)**

*Direct FA Conjugate*

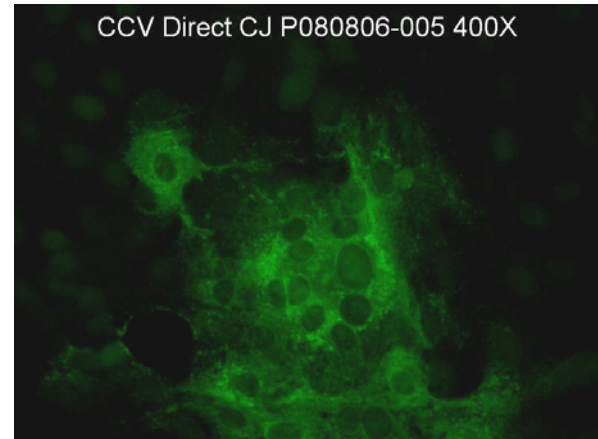
**CATALOG NO.:** 210-21-CCV

**VOLUME:** 10 ml

**LOT:** P080806-005

**EXPIRATION:** 14 August 2010

**VIRUS:** Canine Coronavirus (CCV)



**DESCRIPTION:** Anti-CCV polyclonal antiserum conjugated to fluorescein isothiocyanate. Porcine origin. Ready to use.

**QUALITY CONTROL METHOD:** Direct FA on CCV-infected cell cultures using VMRD, Inc. control slides (catalog no. SLD-FAC-CCV).

**Specific Reaction:** 3-4+ on positive cells; negative on negative cells.

**Other Reactions or Comments:** No background.

**INTENDED USE:** Detects CCV in gut sections or smears, and in cell cultures for identification of isolates. Does not stain other common canine viruses.

**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.

**WARRANTY:** VMRD, Inc. warrants that this product is as described in the quantity and contents stated on the label at the time of delivery to the customer. NO OTHER WARRANTIES, EXPRESS OR IMPLIED, ARE MADE BEYOND THE LABEL DESCRIPTION, INCLUDING WARRANTIES OF MERCHANTABILITY OR FITNESS FOR A PARTICULAR USE. Remedy is limited to replacement of the product or refund of the purchase price. VMRD, Inc. is not liable for property damage, personal injury, or economic loss caused by the product. The information listed in this information sheet is provided for reference only, and should not be substituted for the user's own incoming material quality control.

**RECOMMENDED STAINING PROCEDURE FOR DIRECT FA:**

1. Air dry smears or tissue sections for at least 30 minutes (optimal is overnight) 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, 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 back and edges 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<sub>2</sub>HPO<sub>4</sub> ..... 1.19 gm
- NaH<sub>2</sub>PO<sub>4</sub> ..... 0.22 gm
- NaCl ..... 8.55 gm
- DI/dH<sub>2</sub>O ..... Q.S. to 1 liter

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

- Na<sub>2</sub>CO<sub>3</sub> ..... 11.4 gm
- NaHCO<sub>3</sub> ..... 33.6 gm
- NaCl ..... 8.5 gm
- 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. FA MOUNTING FLUID is made by mixing glycerol and FA rinse buffer, pH 9.0, in equal proportions.