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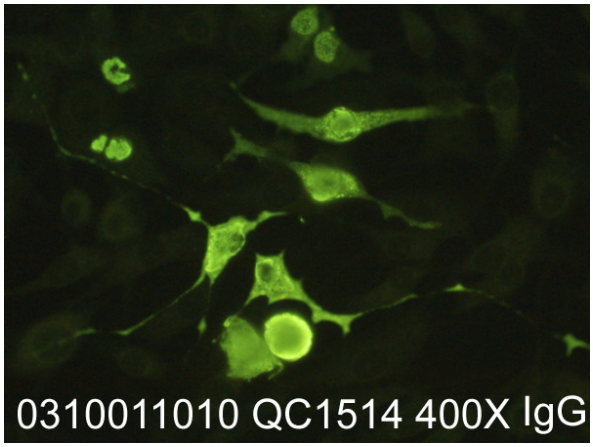
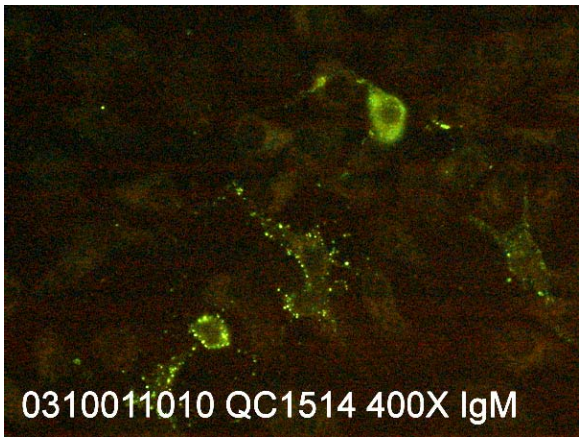
Certificate of Analysis

Canine Parvovirus (CPV)
FA Substrate Slide





CATALOG NO.: 210-88-10-CPV
SIZE: 10 well
LOT: 0310011010-121205
EXPIRATION: 12 December 2005

AGENT: Canine Parvovirus (CPV)
Cell Culture Substrate: Crandell Feline Kidney Cells (CrFK)
Virus Strain: F11/C2-7A

QUALITY CONTROL METHOD: Indirect FA using VMRD CPV IgG Positive Control (211-P-CPV-G); anti-Canine IgG FITC Conjugate (035-10); CPV IgM Positive Control (211-P-CPV-M); anti-Canine IgM FITC Conjugate (036-10).

Specific Reaction: 3-4+ positive IgG reaction; 3+ positive IgM reaction. 5-15 positive cells per high power field.

PATTERN OF FLUORESCENCE: IgG, single cells with diffuse cytoplasmic and nuclear fluorescence. Some degenerate cells with extra cellular antigen. IgM, particulate extra cellular fluorescence.

INTENDED USE: Useful in detecting viral antibody by IFA. May be used to differentiate antibody class (IgG or IgM) with suitable quality fluoresceinated second antibody conjugate.

STABILITY: Foil-pouch sealed slides are stable for at least 6 months when stored below -20°C. Avoid self-defrosting freezers.

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. Screen sera at 1:50 in Special Serum Diluting Buffer (210-94-SB) for IgG testing.

FOR RESEARCH AND INVESTIGATIONAL USE ONLY.

RECOMMENDED STAINING PROCEDURE FOR INDIRECT FA:

1. Warm slide to room temperature before removing from foil pouch.
2. Place 50 µl diluted serum on the designated wells. Dilute serum in serum diluting buffer, pH 7.2 (catalog no. 210-93-SB). [If background is a problem, particularly at low dilutions, use of 10% adult bovine serum diluting buffer is preferable (catalog no. 210-94-SB).]
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. 210-90-RB) 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 50 µl 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 back and edges 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. 210-92-MF) 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 50 µl of 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. 210-90-RB) and then soak for 10 minutes in FA rinse buffer, pH 9.0.
5. Drain slide and dry back and edges 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. 210-92-MF) and view with good quality fluorescence microscope at 100X-250X. Confirmation may be at 400X.

SERUM DILUTING BUFFER (pH 7.2):*

- Na_2HPO_4 . . . . . 1.19 gm
- NaH_2PO_4 . . . . . 0.22 gm
- NaCl . . . . . 8.55 gm
- BSA . . . . . 10.0 gm
- DI/dH_2O . . . . . Q.S. to 1 liter

* This recipe makes 1 liter. If you need less, adjust recipe accordingly. Store at 4°C. Add 0.09% NaN_3 if diluted serum is not going to be used within one week.

4X FA RINSE BUFFER (pH 9.0):

- Na_2CO_3 . . . . . 11.4 gm
- $NaHCO_3$ . . . . . 33.6 gm
- NaCl . . . . . 8.5 gm
- DI/dH_2O . . . . . 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.