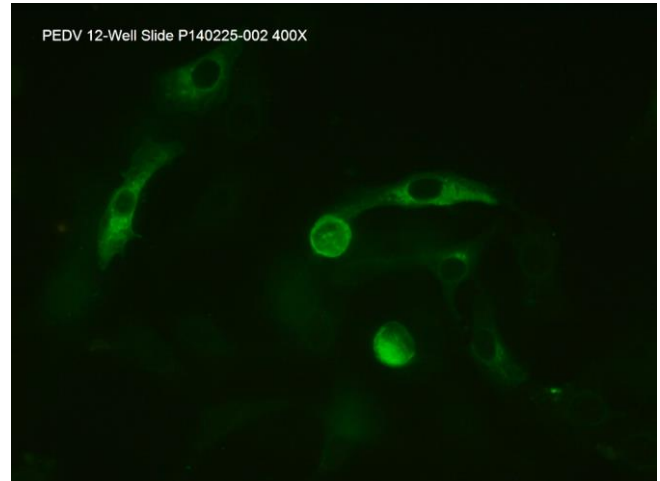


CERTIFICATE OF ANALYSIS

Porcine Epidemic Diarrhea Virus (PEDV)

FA Substrate Slide

Catalog No.:	SLD-IFA-PEDV
Size:	12 Well
Well Capacity:	50 µl
Lot:	P140225-002
Expiration:	17 March 2018
Agent:	Porcine Epidemic Diarrhea Virus (PEDV)
Strain:	PEDV ISU18984IA13
Cell Culture Substrate:	Vero-81



Description:

Wells contain virus-infected cell cultures grown on the surface of Teflon-masked slide. Slides 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 PEDV in-house antiserum and anti-porcine IgG affinity purified conjugate (catalog no. CJ-F-PORG-AP-1ML or 10ML).

Specific Reaction: 3-4+ fluorescence with PEDV antiserum at 1/100, no background. The diluent control was negative. There are 0 to 50 infected cells per high-power field.

Other Comments: NA

Pattern Of Fluorescence:

Primarily granular and diffuse cytoplasmic fluorescence. Mainly single cells with some multinucleated cells.

Intended Use:

Generally used for Indirect FA to detect antibody to PEDV but may also be used as a positive and negative control substrate slide for Direct FA conjugates when applicable.

Storage:

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

References: NA

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. FASDB-100ML) however if high background due to anti-bovine IgG activity is present it may be advisable to use 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 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. 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 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. FAMF-10ML) and view with good quality fluorescence microscope at 100X-250X. Confirmation may be at 400X.

Serum Diluting Buffer (pH 7.2):*

- Na₂HPO₄.....1.19 gm
- NaH₂PO₄.....0.22 gm
- NaCl.....8.55 gm
- BSA.....10.0 gm
- DI/dH₂O.....Q.S. to 1 liter

*This recipe makes 1 liter. If you need less, adjust recipe accordingly. Store at 2-7°C. Add 0.09% NaN₃ if diluted serum is not going to be used within one week.

4X FA Rinse Buffer (pH 9.0):

- Na₂CO₃.....11.4 gm
- NaHCO₃.....33.6 gm
- NaCl.....8.5 gm
- DI/dH₂O.....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.

FOR *IN VITRO* LABORATORY USE ONLY.