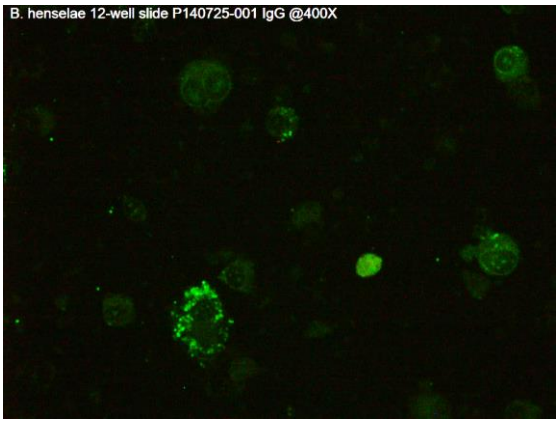
Veterinary Medical
Research & Development

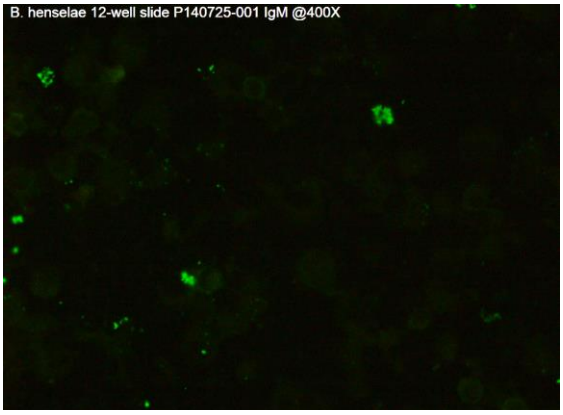
CERTIFICATE OF ANALYSIS

Bartonella henselae

FA Substrate Slide

Catalog No.:	SLD-IFA-BH
Size:	12 Well
Well Capacity:	10 µl
Lot:	P140725-001
Expiration:	28 July 2030
Agent:	*Bartonella henselae*
Strain:	Angu305
Cell Culture Substrate:	DH-82 cells





Description:

Slides contain fixed *Bartonella henselae* in DH-82 cells. Cells are unstained and slides are sealed in moisture-free foil pouches.

Quality Control Method:

Indirect FA using *Bartonella henselae* IgM positive control (catalog no. PC-IFA-BH-M) and anti-feline IgM FITC conjugate (catalog no. CJ-F-FELG-AP-10ML).

Specific Reaction: 3–4+ signal with no background. The diluent control was negative with no background. There are 0 to 3 infected cells per high-power field.

Other Comments: NA

Pattern Of Fluorescence:

Numerous intra-cellular bacteria; both solid and toroidal (donut) fluorescence ranging from 1+ to 4+.

Interpretation Of Results:

Titers of 1/50 (IgG) and greater are considered positive.

Intended Use:

For detection of antibody to *B. henselae* by indirect FA technique. May be used to differentiate antibody class (IgG or IgM) with suitable quality fluoresceinated second antibody conjugates.

Storage:

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

References: NA

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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):*

- 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 2-7°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.