Veterinary Medical
Research & Development

CERTIFICATE OF ANALYSIS

Neospora caninum

FA Negative Control

Catalog No.:	NC-IFA-NC-BOV
Volume:	1 ml
Lot:	P140225-003
Expiration:	10 August 2025
Agent:	*Neospora caninum*

Description:

Bovine serum diluted in PBS, 1% BSA and preserved with 0.09% sodium azide.

Quality Control Method:

Indirect FA using *Neospora caninum* 12-well slide (catalog no. SLD-IFA-NC), bovine positive control (catalog no. PC-IFA-NC-BOV), and anti-bovine $IgG_{1,2}$ FITC affinity purified conjugate (catalog no. CJ-F-BOVG-AP-1ML or 10ML).

Specific Reaction: Trace signal with no background.

Other Comments: NA

Pattern Of Fluorescence:

None with our system.

Intended Use:

As a negative control serum in detection of antibody *to Neospora caninum by indirect* FA technique. This control serum should be used undiluted to demonstrate negative fluorescence, if any.

Storage:

Store at 2-7°C. DO NOT FREEZE! If control becomes cloudy it should be discarded.

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.