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Neospora caninum Antibody Test Kit, cELISA



VMRD's *Neospora* test is a competitive enzyme-linked immunosorbent assay (cELISA) that detects antibodies against *Neospora caninum* in cattle sera. Our competitive ELISA format allows other species to be tested, but validation has been completed only on cattle. An immunodominant surface protein of 65 kDa is captured on the antigen plate using a monoclonal antibody. Another horseradish peroxidase-conjugated monoclonal antibody competes with serum antibodies for a specific epitope on p65. Sensitivity and specificity studies on both species confirm the high accuracy of this kit. In a mass screening of 4323 sera of unknown serologic status only 5% of sera fell within $\pm 5\%$ of the cut-off value, confirming a clear distinction between positive and negative sera bimodal distribution.

VMRD's *Neospora* kit is available in a 2-plate format with breakaway stripwells (and in a 5-plate format with breakaway stripwells by special order).

bovine samples		Reference Assay		
		+	-	Sum
VMRD <i>Neospora</i> cELISA	+	131	4	135
	-	6	319	325
Sum		137	323	460
Sensitivity: 96% • Specificity: 99%				

VMRD cELISA Field Testing, 2001.

KIT CONTENTS

Component	280-2
A Antigen-Coated Plates	2 plates
B Positive Control	3.6 ml
C Negative Control	3.6 ml
D 100X Antibody-Peroxidase Conjugate	300 μ l
E Conjugate Diluting Buffer	30 ml
F 10X Wash Solution Concentrate	120 ml
G Substrate Solution	30 ml
H Stop Solution	30 ml
An insert with Setup Record for recording sample identifications and results.	

Overview of the VMRD *Neospora* Kit Procedure

1	Add 50 μ l of samples and controls into Antigen Plate
2	Incubate 60 minutes at room temperature
3	Wash three times with Wash Solution
4	Add 50 μ l of Conjugate to all wells
5	Incubate Conjugate 20 minutes at room temperature
6	Wash three times with Wash Solution
7	Add 50 μ l of Substrate Solution to all wells
8	Incubate 20 minutes at room temperature
9	Add 50 μ l of Stop Solution to all wells
10	Read at 620-650 nm
Samples causing <30% inhibition are negative. Samples causing $\geq 30\%$ inhibition are positive.	
Formula for calculating % inhibition: $\%I = 100 - [(Sample\ OD / Mean\ Negative\ OD) \times 100]$	
The mean OD of the Negative Control must be ≥ 0.300 and < 2.500. The percent inhibition of the Positive Control must be $\geq 30\%$.	

About Neosporosis

Neosporosis is a recently-described disease that has been identified across the world in various species, including dogs, cattle, sheep, goats, and horses. It is caused by *Neospora caninum*, a protozoan parasite closely related to *Toxoplasma gondii*. Even though the dog can be the definitive host for *Neospora*, it is not known if there are other definitive hosts. No signs of clinical illness are noted in cows that abort due to *Neospora* either prior to the abortion or post-abortion. The aborted fetuses are usually autolyzed with no gross lesions and placentas are not retained. Abortions have been diagnosed in both heifers and cows from three months gestation to term. A majority (78%) of *Neospora* abortions occur between four and six months gestation. This pattern of mid-gestation abortion is distinct from other diagnosed causes of infectious abortion in dairy cattle which tend to occur later in gestation. In dogs, *Neospora* infection causes neuromuscular paralysis. Identification of carrier animals is based upon detection of specific antibody with serological tests while diagnosis of abortions is based upon microscopic examination of the fetus and immunohistochemistry.

Neospora caninum Antibody Test Kit, cELISA

Format	Species	Sample	Sensitivity	Specificity	Assay Time
cELISA	multiple	serum	96%	99%	115 minutes

USDA Licensed Product

Catalog No.	Configuration	Tests
280-2	2 stripwell plates	184
280-5 [†]	5 stripwell plates	460

[†] Custom orders only, with a minimum purchase of 50 kits. Three months lead time required.