

New Product Announcement!



VMRD

P. O. Box 502
Pullman, WA 99163

Phone: (800) 222-8673
FAX: (509) 332-5356
E-mail: order@vmrd.com
techserve@vmrd.com
Web: www.vmrd.com

Equine Infectious Anemia Virus Antibody Test Kit, ELISA

About Equine Infectious Anemia

Equine Infectious Anemia (EIA) is caused by a lentivirus. It produces acute episodes of disease that are interspersed with clinically normal periods. The acute episodes usually last for a few days and are associated with fever, thrombocytopenia, and anemia. In most infected horses, the disease episodes occur with less and less frequency until an inapparent carrier state develops. The infection is life long and, if stressed, inapparent carrier horses may express recurrent viremia and disease. Transmission occurs by transfer of blood from one horse to another by biting insects or contaminated needles and instruments. Transmission is most likely during episodes of clinical disease when the virus titer is highest in the blood, and is least likely during the carrier stage. Unfortunately, it is difficult to know at what stage an infected horse may be and when another episode might occur. It can be diagnosed by detection of antibody to the capsid p26 protein of the virus. This internal viral protein is relatively conserved among EIA virus strains, allowing detection of antibody in virtually all infected horses.

VMRD's **New** Equine Infectious Anemia Virus (EIAV) enzyme-linked immunosorbent assay (ELISA) detects antibodies to EIAV p26 in equine sera. EIAV-specific antibodies bind to recombinant p26 antigen when sera are added to the test wells. Unbound antibody is washed away and antigen-bound antibody is detected by the addition of horseradish-peroxidase (HRP)-conjugated

p26 antigen. Unbound conjugate is washed away and, with the addition of substrate solution, wells containing antibody turn blue. When stop solution is added, positive wells turn yellow and negative wells remain clear. Test results may be calculated from optical density values reported by a microplate absorbance reader or determined visually by comparing color development intensity of sample wells with that of the positive control.

EIAV ELISA

VMRD's EIAV ELISA is rapid and convenient—only 35 minutes total incubation time, no sample dilution, and only two washes—yet it is highly specific and sensitive.

Field Testing Data		Reference Assay		
		+	-	Sum
VMRD EIA ELISA	+	122	0	122
	-	0	421	421
	Sum	122	421	543

Sensitivity: 100%* • Specificity: 100%*

In-House Testing

1628 AGID-negative samples tested negative with VMRD's ELISA. It correctly classified all samples in the 2001, 2002, 2003, 2004, and the 2005 USDA CVB EIAV check sets. On a panel of 10 "weak samples," VMRD's assay correlated 100% with the consensus of three USDA-licensed EIAV ELISAs.

Field Testing

In field trails conducted by three independent laboratories on a total of 543 samples, VMRD's new ELISA had 100% correlation with the various reference assays used. As a test of robustness, a tester independency panel consisting of 25 samples—12 positive and 13 negative—was distributed to the participating laboratories. All were in agreement on all samples with the exception of one laboratory that called one negative sample positive. VMRD's ELISA sensitivity is comparable or superior to other USDA-licensed ELISAs on titrations of positive samples and in detection of "weak samples." VMRD's kit contains no thimerosal and generates no hazardous waste.



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Cat. No.	Species	Sample	Sensitivity*	Specificity*	Incubation Time	Configuration	Tests
290-1	equine	serum	100%	100%	35 minutes	1 stripwell plate	94
290-5						5 stripwell plates	470

VMRD's ELISA in a side-by-side comparison with three other commercially available EIAV ELISAs

Known Positive Samples	VMRD ELISA	ELISA X	ELISA Y	ELISA Z
A @ Neat	+	+	+	+
1:100	+	-	+	+
1:200	+	-	+	+
1:400	+	-	+	+
1:800	+	-	+	+
1:1600	-	-	-	-
B @ Neat	+	+	+	+
1:64	+	+	+	+
1:128	+	-	+	+
1:256	+	-	+	+
1:512	+	-	-	+
C @ Neat	+	+	+	+
1:8	+	+	+	+
1:16	+	+	+	+
1:32	+	-	-	-
D (weak +)	+	+	+	+
E	+	+	-	+
F (weak +)	+	+	+	+
G (weak +)	+	+	+	+
H (weak +)	+	+	+	+

Overview of the EIAV ELISA Kit Procedure

- 1 Place 50 µl of samples and controls into wells of Antigen Plate
- 2 Incubate 10 minutes at room temperature
- 3 Wash once with Wash Solution
- 4 Add 50 µl of Conjugate
- 5 Incubate 10 minutes at room temperature
- 6 Wash four times with Wash Solution
- 7 Add 50 µl of Substrate Solution
- 8 Incubate 15 minutes at room temperature
- 9 Add 50 µl of Stop Solution to all wells
- 10 Read at 450 nm or by eye

Samples are positive if they produce an OD greater than or equal to the mean of the positive control.

Samples are negative if they produce an OD less than the mean of the positive control.

For the test to be valid, the OD of the Positive Control should be greater than or equal to 1.5 times the OD of the Negative Control. The OD of the Negative Control should be less than or equal to 0.15.

For the test to be valid when reading by eye, the Positive Control should have visible yellow color and the Negative Control should have no or faint visible color that is less than the Positive Control.

Samples producing positive test results are to be sent in to the National Veterinary Services Laboratories (NVSL) for verification.



VMRD's EIAV ELISA, like all VMRD ELISAs, is supported by VMRD's new **ELISAWare™** microplate reading software. **ELISAWare™** works with a variety of microplate reading instruments and operating systems, so there's a good chance that you can use it with your existing hardware. You can find more information about **ELISAWare™** at www.vmrd.com/elisaware



VMRD's ELISA generates no hazardous waste!

VMRD's EIAV ELISA does not contain Thimerosal, simplifying disposal. There is no extra cost associated with storage or disposal of hazardous products.

*Sensitivity and Specificity in Perspective

Relative sensitivity and specificity values are calculated from data generated by diagnostic laboratory field testing. These values are provided as guidelines only and should not be construed as the absolute sensitivity and specificity of the test in question for any population subset.