

Porcine Reproductive and Respiratory Syndrome Virus (PRRSV) Antibodies and ELISA Kits

PRRSV is a highly contagious disease and most economically devastating disease in the swine industry. It affects millions of pigs in the world. Rapid diseases detection and control is necessary to preserve billions of dollars of animal trade. As a result of PRRS infection, antibodies to several **structural proteins** (Nucleocapsid protein or NP etc.) and **non-structural proteins (NSPs)** are induced. However, NP antibodies are generated at low levels and later in infection, whereas NSP antibodies can be detected much sooner (7-14 weeks post infection or DPI) and stay at higher levels for 6-9 months. Therefore, NSP-antibody based PRRS detection is more sensitive and robust diagnostic tests. Until now only low sensitivity PRRS Type I+II NP IgG commercial ELISA are available. ADI now offers the most sensitive, simple, and rapid ELISA kits for the **detection of PRRS NP antibodies** that can be used for assessing vaccine status and/or infection. **PRRSV NSP antibodies ELISA kits** are used for assessing natural infection (**DIVA tests**). ADI ELISA produced results in 105 min at room temp and has a high sensitivity of 1 ng PRRS antibody.

PRRSV mouse antibody ELISA kits are suitable for PRRSV vaccine efficacy testing.

Assay Procedure: Arrange required number of strips on the plate.

- Step 1. Add **100 µl of pre-diluted Controls** (-Ve, Cut-off and +ve) and samples (diluted 1:50 or higher) into respective wells. Mix gently and **incubate at room temp for 60 mins** (25-28°C; no shaking necessary).
- Step 2. **Aspirate well contents and wash 3X** with wash buffer. Add **100 µl of supplied antibody-HRP Conjugate** (anti-Pig IgA or IgM or IgG) into all wells; mix gently and **incubate at RT for 30mins**.
- Step 3. **Aspirate or wash 5x** with wash buffer. Tap plates over paper towels. Add **100 µl of TMB Substrate**. Mix gently and **Incubate for 15 min** at RT. **Blue color** develops in positive wells.
- Step 4. Add **100 µl of stop solution** into each well and mix gently (blue color turns yellow). **Measure yellow color at 450 nm**. Results are compared to Cut-off control and expressed as +ve and -ve or antibody values determined from Antibody standard curve and expressed as U/ml.

Calculation of Results

Results can be expressed as simple -ve and +ve or PRRS antibody concn (U/ml) determined from standard curve.

Typical ELISA Results

| PRRS ELISA | PRRS I+II NSP IgG (U/ml) | Net A450 |
|-----------------------|--------------------------|----------|
| -ve control | 1 | 0.101 |
| Cut-off control | 10 | 0.300 |
| Positive control | 30 | 0.610 |
| High positive control | 100 | 1.230 |

Fig. 1. Typical results are shown for PRRS Type I+II Combo. These kits are suitable for assessing vaccination (NP/NSP ELISA) or infection (NSP ELISA).

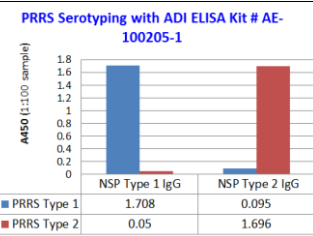
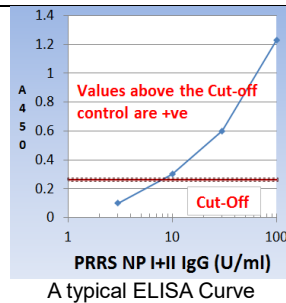


Fig. 2. Sera from experimentally infected pigs with PRRS Type 1 or Type 2 (27 dpi) were tested at 1:100 dilution on ADI PRRS NSP serotyping ELISA kit. Serotypes were clearly identified.

PRRS ELISA Kits Ordering Information

| PRRSV Type | ELISA kit Description | PRRSV IgG |
|---|---|-------------|
| Type 1 (European) | Recombivirus Mouse anti-PRRS NP Type 1 IgG ELISA kit (96 wells) | RV-400105-1 |
| Type 2 (North American) | Recombivirus Mouse anti-PRRS NP Type 2 IgG ELISA kit (96 wells) | RV-400115-1 |
| Type 1+2 Combo | Recombivirus Mouse anti-PRRS NP Type 1+2 IgG, Combo ELISA kit (1x 96 or 5x96 tests) (detects I+II as a mix) | RV-400125-1 |
| Type 1 (European) | Recombivirus Mouse anti-PRRS NSP Type 1 IgG ELISA kit (96 wells) for Serotyping of type 1 DIVA Tests | RV-400135-1 |
| Type 2 (North American) | Recombivirus Mouse anti-PRRS NSP Type 2 IgG ELISA kit (96 wells) for Serotyping of type 2 DIVA Tests | RV-400145-1 |
| Type 1+2 Combo | Recombivirus Mouse anti-PRRS NSP Type 1+2 IgG, Combo ELISA kit (1x 96 or 5x96 tests) (detects I+II as a mix) DIVA Tests | RV-400155-1 |
| Recombinant PRRSV antigen coated plates for ELISA | Recombinant PRRSV NP Type 1 antigen coated plates for ELISA (5x96 tests) | RV-400106-1 |
| | Recombinant PRRSV NP Type 2 antigen coated plates for ELISA (5x96 tests) | RV-400116-1 |
| | Recombinant PRRSV NP Type 1+2 antigen coated plates for ELISA (5x96 tests) | RV-400126-1 |
| | Recombinant PRRSV NSP Type 1 antigen coated plates for ELISA (5x96 tests) | RV-400136-1 |
| | Recombinant PRRSV NSP Type 2 antigen coated plates for ELISA (5x96 tests) | RV-400146-1 |
| | Recombinant PRRSV NSP Type 1+2 antigen coated plates for ELISA (5x96 tests) | RV-400156-1 |

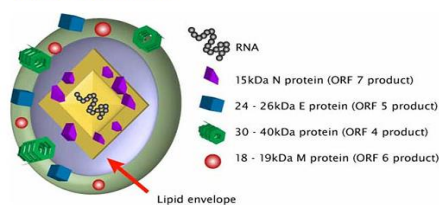
PRRSV Recombinant Proteins, Antibodies, and Serum Controls

| Type | Catalog# | Product Description | Product Type |
|------------------------------------|--------------|---|------------------------|
| PRRSV Type 1 (European) | PRSNP11-PNC | Porcine reproductive and Respiratory syndrome virus 1 nuclear protein (PRRSV1-NP) antibody negative control serum | Disease serum, Porcine |
| | PRSNP12-PPC | Porcine reproductive and Respiratory syndrome virus 1 nuclear protein (PRRSV1-NP) antibody positive control serum | Disease serum, Porcine |
| | PRSNP15-R-10 | Recombinant (E.coli) Porcine reproductive and respiratory syndrome virus 1 nuclear protein (PRRSV1-NP) (His tag, >95%) | Recombinant protein |
| | PRSNP11-S | Anti-Porcine reproductive and Respiratory syndrome virus 1 nuclear protein (PRRSV1-NP) antiserum | Antiserum |
| | PRSNP13-PNC | Porcine reproductive and Respiratory syndrome virus 1 nonstructural protein1 (PRRSV1-NSP1) antibody -ve control serum | Disease serum, Porcine |
| | PRSNP14-PPC | Porcine reproductive and Respiratory syndrome virus 1 nonstructural protein1 (PRRSV1-NSP1) antibody +ve control serum | Disease serum, Porcine |
| | PRSNP16-R-10 | Recombinant (E.coli) PRRSV1 nonstructural protein (PRRSV1-NSP1) (his tag >95%) | Recombinant protein |
| | PRSNP15-S | Anti-Porcine reproductive and Respiratory syndrome virus 1 nonstructural protein (PRRSV1-NSP) antiserum | Antiserum |
| PRRSV Type 2 (American) | PRSNP21-PNC | Porcine reproductive and Respiratory syndrome virus 2 nuclear protein1 (PRRSV2-NP1) antibody negative control serum | Disease serum, Porcine |
| | PRSNP22-PPC | Porcine reproductive and Respiratory syndrome virus 2 nuclear protein1 (PRRSV2-NP1) antibody positive control serum | Disease serum, Porcine |
| | PRSNP25-R-10 | Recombinant (E.coli) Porcine reproductive and respiratory syndrome virus 2 nuclear protein (PRRSV2-NP) (his tag, >95%) | Recombinant protein |
| | PRSNP21-S | Anti-Porcine reproductive and Respiratory syndrome virus 2 nuclear protein (PRRSV2-NP) antiserum | Antiserum |
| | PRSNP23-PNC | Porcine reproductive and Respiratory syndrome virus 2 nonstructural protein1 (PRRSV2-NSP) antibody -ve control serum | Disease serum, Porcine |
| | PRSNP24-PPC | Porcine reproductive and Respiratory syndrome virus 2 nonstructural protein1 (PRRSV2-NSP) antibody positive control serum | Disease serum, Porcine |
| | PRSNP26-R-10 | Recombinant (E.coli) PRRSV2 nonstructural protein (PRRSV2-NSP) (his tag, 30 kDa) purified | Recombinant protein |
| | PRSNP22-S | Anti-Porcine reproductive and Respiratory syndrome virus 2 nonstructural protein (PRRSV2-NSP) antiserum | Antiserum |

Porcine Reproductive and Respiratory Syndrome Virus (PRRSV) –General Information

Porcine Reproductive and Respiratory Syndrome (PRRS) is a disease found in swine farms worldwide and it is characterized by reproductive failure such as late-term abortions in sows and by respiratory illness and mortality in young pigs. PRRS is caused by porcine reproductive and respiratory syndrome virus (**PRRSV**), a small, enveloped, positive-stranded RNA virus. It is a member of the family Arteriviridae, which includes Equine arteritis virus, Lactate dehydrogenase elevating virus of mice and Simian hemorrhagic fever virus in the order Nidovirales. The pig (*Sus scrofa*), whether domestic or feral, is the only species known to be **naturally susceptible to PRRS disease**. The continuous circulation of the virus among the pig population causes severe economic loss for the swine industry.

Virus structure



PRRSV can be differentiated into two genotypes: **Type 1 (European strains) and Type 2 (North American strains)** that share ~ 55-60%

nucleotide sequence identity. The extensive antigenic and genetic variations among field strains of PRRSV are largely responsible for the poor cross-protection of the current vaccines against heterologous strains. The **PRRSV genome** is ~ **15 kb** and encodes the **non-structural proteins (nsp1-12)** that play a

key role in viral replication and transcription. The virion contains three major structural proteins, an **envelope glycoprotein** (ORF5, GP5 ~25 kda), unglycosylated **membrane protein M** (ORF6, ~18 kda), and **nucleoprotein or nucleocapsid** (ORF7, N ~15 Kda). ORF7/NP is among the most antigenic proteins of PRRS upon infection.

Diagnosis of PRRSV is by **virus isolation (VI)**, detection of PRRS antigen by fluorescent antibody tests (FAT) or immunohistochemistry (IHC), or detection of PRRS by **PCR**. Serology tests for PRRS antibody like indirect immunofluorescent antibody (**IFA**), serum neutralization, and enzyme-linked immunosorbent assay (**ELISA**) have been used. Commercial ELISA kits based on PRRSV antigens from type 1+2 (Combo) is widely used for the diagnosis of PRRS. There are no good options in using **PRRS DIVA** test due to inherent issues in using appropriate PRRS vaccines that will allow proper testing.

All currently approved **PRRSV Vaccines** include either **modified-live virus (MLV) vaccine and or killed virus (KV) vaccine**. PRRS MLV vaccine is well recognized for its protective efficacy against PRRSV that are genetically homologous to the vaccine virus. It is of concern, however, for its immunogenicity, cross protective efficacy and safety. PRRS KV vaccine, on the other hand, is well known for its safety, but it only confers limited protection.

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