HIGHLY PATHOGENIC PORCINE REPRODUCTIVE AND RESPIRATORY SYNDROME VIRUS

Etiology

• Porcine reproductive and respiratory syndrome virus (PRRSV) is an enveloped, single-stranded RNA virus within the family Arteriviridae. Other members of the family include equine arteritis virus, lactate dehydrogenase-elevating virus (LDV), and simian hemorrhagic fever virus.
• PRRSV emerged almost simultaneously in the late 1980s in the U.S. and Germany, causing severe reproductive and respiratory disease in pigs.
• Highly pathogenic PRRSV (HP-PRRSV) was first described in China in 2006, following an outbreak in which 2 million pigs were affected and 400,000 died. The virus originated from a type 2 (North American) PRRSV already circulating in China. New HP-PRRSVs continue to emerge and highly pathogenic forms of the virus are now dominant in China.

Cleaning and Disinfection

• PRRSV is stable over a pH range of 6.5–7.5, and the virus can survive for years at temperatures of -70°C to -20°C (-94°F to -4°F). Experimentally, PRRSV has been shown to survive in pork under various conditions, but the introduction of PRRSV through importation of pork has been deemed unlikely.
• PRRSV is susceptible to phenol, formaldehyde and many common disinfectants such as bleach, iodine, and quaternary ammonium compounds. No information is available regarding survival or disinfection specifically for HP-PRRSV.

Epidemiology

• Pigs are the only known natural hosts for PRRSV and HP-PRRSV; there is no evidence that PRRSV or HP-PRRSV are zoonotic.
• HP-PRRSV causes higher morbidity and mortality compared to PRRSV. In the original 2006 Chinese outbreak, morbidity rates ranging from 50–100% were noted. An overall mortality rate of nearly 20% was observed, with up to 100% mortality in individual herds. Mortality rates were highest in suckling pigs (100%), followed by nursery pigs (70%), finishers (20%), and pregnant sows (10%).
• PRRSV has become an economically devastating disease of pigs that is found in many, but not all, swine-producing areas of the world. HP-PRRSV—which has not been described in the U.S.—has been detected in China, Vietnam, the Lao People’s Democratic Republic, Thailand, Cambodia, and India.
Transmission
- Infected pigs shed PRRSV in respiratory secretions, saliva, urine, semen, mammary secretions, and occasionally feces.
- PRRSV is transmitted directly via intranasal, oral, intrauterine/transplacental, and vaginal routes. Experimental inoculation via the intramuscular route is effective in reproducing the disease. The virus can be spread through breaks in the skin such as bites, cuts, or scrapes, as well as husbandry practices like ear notching and tail docking. Aerosol transmission has been demonstrated and may serve as an important means of virus spread between farms/herds. Fomites and mechanical insect vectors have also been implicated in PRRSV transmission.
- It is not known how transmission of HP-PRRSV differs from PRRSV, if at all.
- PRRSV endemicity in a herd is driven by prolonged infection in carrier animals (more than 200 days) and the continued presence of susceptible animals (usually suckling piglets) in a herd. Vertical transmission is a major contributor to within-herd PRRSV transmission (e.g., pigs that are viremic at birth), as is comingling of infected and susceptible pigs.

Infection in Swine/Pathogenesis
- PRRSV replicates mostly in the lymphoid tissues and lungs, although HP-PRRSV isolates may have a broader tissue tropism compared to PRRSV.
- The clinical presentation is variable. In breeding animals, anorexia, fever, lethargy, depression, respiratory distress, and vomiting can be seen, as well as cyanosis of the ears, abdomen, and vulva. Transplacental transmission results in reproductive failure (stillborn, autolysed, and/or mummified fetuses) or birth of viremic piglets. In young, growing, and finishing pigs, acute viremia is followed by respiratory disease including pneumonia, sneezing and expiratory dyspnea. Fever, lethargy and depression can also be seen.
- HP-PRRSV causes similar clinical manifestations; however, infections with highly virulent strains are characterized by increased disease severity. Additionally, neurological signs have been associated with HP-PRRSV, as well as an erythematous blanching rash. Unlike PRRSV, HP-PRRSV causes clinical disease and death in all ages, including adult pigs and pregnant sows.

Diagnosis
- PRRSV can be detected via direct fluorescent antibody, immunohistochemistry, or nucleic acid-based assays. Several variations on the reverse transcriptase polymerase chain reaction (RT-PCR) assay have been developed. Loop-mediated isothermal amplification (LAMP) tests and rapid point-of-care immunochromatographic strip tests have been described, but are not widely available or routinely used in the U.S.
- Although traditional RT-PCR assays cannot distinguish PRRSV from HP-PRRSV, newer RT-PCR variations available in China are capable of differentiation. These tests are not yet widely available in the U.S.
- Serological tests for PRRSV antibody include indirect fluorescent antibody and an enzyme linked immunosorbent assay (ELISA) targeting the nucleocapsid protein.
- Samples suitable for virus detection include serum, lung, lung lavage, lymph nodes, spleen, heart, kidney, thymus, and tonsil (especially in chronically infected pigs). PRRSV can also be detected in oral fluids.

Immunity
- PRRSV is known to suppress innate immunity in infected pigs. Although PRRSV can persist long-term in carrier pigs, it seems that most infected pigs become immune and viral shedding usually stops sometime before 60 days post-infection (DPI).
• PRRSV vaccination may moderate clinical signs and reduce virus shedding. Inactivated and modified live vaccine (MLV) formulations are available. However, efficacy of PRRSV vaccination is unpredictable because the virus mutates rapidly and many vaccines perform poorly following heterologous challenge.
• Infection with one PRRSV strain does not prevent infection with a heterologous strain. There is evidence demonstrating that herds can be simultaneously infected with more than one PRRSV.

Prevention and Control
• Methods to prevent the entry of PRRSV into a naïve swine herd include quarantine and testing of new breeding animals; sanitation and drying of transport/supply vehicles; shower-in, shower-out or use of Danish entry systems; and implementation of insect control program (e.g., screening, habitat management, and insecticide use). Air filtration systems have been shown to reduce the risk of PRRSV transmission.
• In PRRS-endemic herds, control programs can be implemented to limit adverse effects of the disease. Best control strategies will vary depending on specific farm situations; however, PRRS control programs often involve a combination of gilt acclimatization, restricted entry of incoming breeding stock, restricted cross-fostering, euthanasia of affected pigs, all-in/all-out pig flow, partial or total herd depopulation and repopulation, segregated early weaning, test and removal, and herd closure.
• Vaccination has been the main control measure implemented in China against HP-PRRSV.

Gaps in Preparedness
• PRRSVs have been economically devastating for swine producers in many parts of the world, and new PRRSV variants—causing even higher high morbidity and mortality—continue to emerge in Asia. Potential entry routes into the U.S. should be investigated. Strict biosecurity measures should be in place to prevent an HP-PRRSV incursion.
• To advance our understanding of PRRSV, future studies on basic viral biology, pathogenesis, host genomics, viral genomics, and immunology must continue.
• Prolonged carriage in pigs makes PRRS elimination difficult. Control programs are hampered by the virus’ rapid evolution and lack of heterologous protection by current vaccines. New vaccine technologies must be developed to enhance cross-protective immunity and protection. Additionally, diagnostic tests that can differentiate infected from vaccinated animals (DIVA) are urgently needed. Improved vaccines and diagnostics are critical for successful control of PRRSV.
Porcine reproductive and respiratory syndrome virus (PRRSV) is an enveloped, single-stranded RNA virus within the family *Arteriviridae*. Only one genus, *Arterivirus*, has been recognized. Other members of the family include equine arteritis virus, lactate dehydrogenase-elevating virus (LDV), and simian hemorrhagic fever virus. PRRSV emerged almost simultaneously in the late 1980s in the U.S. and Germany, causing severe reproductive and respiratory disease in pigs. Since then, PRRSV has become an economically devastating disease of pigs that is found in many, but not all, swine-producing areas of the world. PRRSV is discussed in this document mainly to provide background information; highly pathogenic PRRSV (HP-PRRSV), which has not been detected in the U.S. to date, is the focus of this review.

PRRSV has circulated in China since the mid-1990s. Strains with increased pathogenicity, HP-PRRSV, were first described in China in 2006 following an outbreak in which 2 million pigs were affected and 400,000 died. HP-PRRSV originated from a type 2 (North American) PRRSV already circulating in China. New variants continue to emerge and highly pathogenic forms of the virus are now dominant in China. HP-PRRSV has also been detected in Vietnam, the Lao People’s Democratic Republic, Thailand, Cambodia, and India.

PRRSV is stable over a pH range of 6.5–7.5, and the virus can survive for years at temperatures of -70°C to -20°C (−94°F to −4°F). Experimentally, PRRSV has been shown to survive in pork under various conditions, but the introduction of PRRSV through importation of pork has been deemed unlikely. PRRSV is susceptible to phenol, formaldehyde and many common disinfectants such as bleach, iodine, and quaternary ammonium compounds. No information is available regarding survival or disinfection specifically for HP-PRRSV.

Pigs are the only natural hosts for PRRSV. Natural HP-PRRSV infection has been reported in domestic swine and hybrid wild pigs. Wild pigs have been infected experimentally. There is no evidence that PRRSV or HP-PRRSV are zoonotic. HP-PRRSVs cause higher morbidity and mortality compared to PRRSV. In the original 2006 Chinese outbreak, morbidity rates of 50–100% were reported. An overall mortality rate of nearly 20% was observed, with up to 100% mortality in individual herds. Mortality rates were highest in suckling pigs (100%), followed by nursery pigs (70%), finishers (20%), and pregnant sows (10%).

Infected pigs shed PRRSV in respiratory secretions, saliva, urine, semen, mammary secretions, and occasionally feces. PRRSV is transmitted directly via intranasal, oral, intrauterine/transplacental, and vaginal routes. Additionally, intramuscular inoculation has been successfully used to reproduce the disease under experimental conditions. The virus can be spread through breaks in the skin such as bites, cuts, or scrapes, as well as husbandry practices like ear notching and tail docking. Aerosol transmission has been demonstrated and can spread the virus between farms/herds. Fomites and mechanical insect vectors have also been implicated in PRRSV transmission. It is not known how transmission of HP-PRRSV differs from PRRSV, if at all. PRRSV endemicity in a herd is driven by prolonged infection in carrier animals (more than 200 days) and the presence of susceptible animals (usually suckling piglets) in a herd. Vertical transmission is a major contributor to within-herd PRRSV transmission (via birth of viremic piglets), as is comingling of infected and susceptible pigs.

PRRSV replicates mostly in the lymphoid tissues and lungs, although HP-PRRSV isolates may have a broader tissue tropism. The clinical presentation is variable. In breeding animals, anorexia, fever, lethargy, depression, respiratory distress, and vomiting can be seen, as well as cyanosis of the ears, abdomen, and vulva. Transplacental transmission results in reproductive failure (stillborn, autolized, and/or mummified fetuses) or birth of viremic piglets. In young, growing, and finishing pigs, acute
viremia is followed by respiratory disease. In addition to pneumonia, sneezing and expiratory dyspnea, fever, lethargy, and depression may occur. HP-PRRSV causes similar disease but of increased severity. In addition, neurological signs have been associated with HP-PRRSV, as well as an erythematous blanching rash. Unlike PRRSV, HP-PRRSV causes clinical disease and death in pigs of all ages, including adult pigs and pregnant sows. Severe multiorgan damage can be caused by HP-PRRSV and lung lesions are particularly common though not pathognomonic. Frequent viral co-infections can complicate the interpretation of clinical signs and lesions in PRRSV-infected pigs.

PRRSV can be detected via direct fluorescent antibody, immunohistochemistry, or nucleic acid-based assays. Loop-mediated isothermal amplification (LAMP) tests have been developed as well as immunochromatographic strip tests for PRRSV detection; however, they are not widely available or routinely used in the U.S. Although traditional RT-PCR assays cannot distinguish PRRSV from HP-PRRSV, newer RT-PCR assays available in China are capable of differentiation. Serological tests for PRRSV antibody include indirect fluorescent antibody and an enzyme linked immunosorbent assay (ELISA) targeting the nucleocapsid protein. Samples suitable for virus detection include serum, lung, lung lavage, lymph nodes, spleen, heart, kidney, thymus, and tonsil (especially in chronically infected pigs). PRRSV can also be detected in oral fluids.

PRRSV is known to suppress innate immunity in infected pigs. Although PRRSV can persist long-term in carrier pigs, it seems that most infected pigs become immune and viral shedding usually stops sometime before 60 days post-infection (DPI). PRRSV vaccination may moderate clinical signs and reduce virus shedding. Inactivated and modified live vaccine (MLV) formulations are available. However, efficacy of PRRSV vaccination is unpredictable because the virus mutates rapidly and many vaccines perform poorly against heterologous challenge. Infection with one PRRSV strain does not prevent infection with a heterologous strain. Herds can be simultaneously infected with more than one PRRSV.

Methods to prevent the entry of PRRSV into a naïve swine herd include quarantine and testing of newly introduced animals; sanitation and drying of transport/supply vehicles; shower-in, shower-out or use of Danish entry systems; and implementation of insect control programs (e.g., screening, habitat management, and insecticide use). Air filtration systems have been shown to reduce the risk of PRRSV transmission. In PRRS-endemic herds, control programs can be implemented to limit adverse effects of the disease. Best control strategies will vary depending on specific farm situations; however, PRRS control programs often involve a combination of gilt acclimatization, restricted entry of breeding stock, restricted cross-fostering, euthanasia of affected pigs, use of all-in/all-out pig flow, partial or total herd depopulation and repopulation, segregated early weaning, test and removal, and herd closure. Vaccination has been the main control measure implemented in China against HP-PRRSV.

PRRSVs have been economically devastating for swine producers in many parts of the world, and new PRRSV variants—causing even higher morbidity and mortality rates—continue to emerge in Asia. Potential entry routes into the U.S. should be investigated. Strict biosecurity measures should be in place to prevent an HP-PRRSV incursion. To advance our understanding of PRRSV, future studies on basic viral biology, pathogenesis, host genomics, viral genomics, and immunology must continue. Although PRRSV vaccines are available, control programs are hampered by the virus’ rapid evolution and deficient heterologous coverage. New vaccine technologies must be developed to enhance cross-protective immunity and protection. Prolonged carriage in pigs impedes elimination efforts, and diagnostic tests that can differentiate infected from vaccinated animals (DIVA) are urgently needed. Improved vaccines and diagnostics are critical components in a PRRSV control strategies.
LITERATURE REVIEW

1. Etiology

1.1 Key Characteristics
Porcine reproductive and respiratory syndrome virus (PRRSV) is an enveloped virus within the family Arteriviridae. Only one genus, Arterivirus, has been recognized.1 Arteriviruses are pleomorphic with a linear, positive-sense, single-stranded RNA genome.1 Other members of the family include equine arteritis virus, lactate dehydrogenase-elevating virus (LDV), and simian hemorrhagic fever virus.

1.1.1 PRRSV
In the late 1980s, an unknown syndrome emerged in U.S. pigs causing severe reproductive and respiratory disease.2,3 Around the same time a similar syndrome was reported in Germany.4 Initially described as ‘mystery swine disease’, the disease was also known as swine infertility and respiratory syndrome (SIRS), porcine epidemic abortion and respiratory syndrome (PEARS), and blue-eared pig disease. Later named porcine reproductive and respiratory syndrome (PRRS), a viral etiology was confirmed in 1991.5 The origin of PRRSVs remains unknown. However, the virus was likely present in swine long before the ‘original’ epidemics occurred.8 Phylogenetic analysis suggests that PRRSV evolved from a common ancestor with LDV.7

Through much of the world, PRRS has become an economically devastating disease of swine. In the U.S., PRRS costs the swine industry an estimated $664 million dollars annually.8 PRRSV is discussed in this document mainly to provide background information; highly pathogenic PRRSV (HP-PRRSV), which has not been detected in the U.S. to date, is the focus of this review.

1.1.2 Highly Pathogenic PRRSV
PRRSV has circulated in China since the mid-1990s.9 However, ‘swine high fever disease’ was first described in China in 2006, affecting more than 2 million pigs and causing 400,000 deaths over a three-month period.10 By 2007, the epidemic became widespread.11 A new PRRSV variant, known as HP-PRRSV, was isolated and confirmed as the cause.10,12,13 A second major outbreak occurred from 2009–201014, and since then HP-PRRSV has continued to circulate in China where it has become the dominant type.15,16 The virus has also spread to Southeast Asia,17–20 likely through multiple introductions.21 Since 2006, HP-PRRSV variants have continued to emerge. Details are provided in section 1.2.2.

1.2 Strain Variability

1.2.1 PRRSV
The PRRSV genome consists of about 15,000 nucleotides containing 11 open reading frames (ORFs).22 ORFs 1a and 1b encode two nonstructural proteins (NSPs), pp1a and pp1ab, that are further processed into 14 nonstructural proteins (NSPs).22 ORFs 2–7 encode eight structural proteins, including the major envelope proteins GP5 and M, the nucleocapsid protein (N), and the minor envelope proteins E, GP2a, GP3, GP4, and ORF5a.22 ORF5 and NSP2 are the most variable genes in the PRRSV genome.

Based on sequencing of ORF5, which encodes the major envelope glycoprotein GP5, two PRRS genotypes have been identified. Type 1, also known as the European type, is represented by the prototype strain Lelystad virus. Type 2, the North American type, is represented by prototype strain ATCC VR2232.23 Diversity is high both within and between type 1 and type 2 PRRSVs, where only 60% nucleotide homology has been observed.1 In an individual animal, PRRSV exists as a quasispecies (a ‘cloud of mutationally distinct genomes’).23

Type 2 (North American) viruses predominate in China; however, there is evidence that type 1 (European) isolates have also been present since at least 2011.24,25 Although PRRSV strains with increased virulence were reported in the U.S. in 2014 (i.e., PRRSV 1-7-4), these strains were not
classified as HP-PRRSV, having originated from existing wild-type midwestern PRRSVs, not from Chinese isolates.26,27

1.2.2 Highly Pathogenic PRRSV

1.2.2.1 Genetic Markers of HP-PRRSV

Whole genome sequencing of HP-PRRSVs isolated from China in 2006 showed that all were type 2 (North American) viruses10 originating from a PRRSV already present in the country.28 A hypervariable region was identified in NSP2 containing a discontinuous deletion of 30 amino acids (aa). Specifically, a conserved leucine was absent at position 482, and a continuous deletion of 29 aa was observed from positions 534–562.10 These specific changes in NSP2 have become a genetic marker of HP-PRRSV.12,15,16,29-35 However, other NSP2 polymorphisms have previously been identified in type 2 PRRSVs.36,37 Sequence changes in the highly variable GP5 are also a common finding in PRRSVs including highly pathogenic strains.15,29,30,32,33,35,38-41

Since 2006, HP-PRRSV variants have continued to emerge in China. Phylogenetic analysis showed that 2009–2010 outbreak strains were likely a result of natural recombination.42 Viruses from Southern China collected from 2010–2013 contained a unique aa deletion in NSP2, which identified them as a new subgenotype of HP-PRRSV.29 From 2013–2014, a recombinant of a recently introduced and now prevalent North American virus (NADC30-like)43 and HP-PRRSV emerged in China.44 Sequence analysis of HP-PRRSVs collected in Southern China from 2014–2015 identified isolates closely related to NADC30, as well as viruses belonging to new subgenotypes based on heterogeneity in GP5.45

Other genetic variations seen in emerging HP-PRRSVs have involved GP3, a minor structural protein essential for virus infectivity,22,32,33; ORF5a,32,33 a minor structural protein essential for virus viability;22,47 Sequencing of ORF7, which encodes the N protein, has also been useful in phylogenetic analysis.48 Because PRRSVs evolve rapidly, and recombination is common, the viral population is extremely genetically and antigenically diverse.22 It is likely that surveillance will continue to identify new variants of HP-PRRSV.

1.2.2.2 Pathogenicity of HP-PRRSV

Variations in NSP2 are not related to increased virulence associated with the HP-PRRSV.49,50 Evidence has suggested that the NSP9 and 10 coding regions together contribute to the increased pathogenicity of HP-PRRSV.51 Virulence determinants are also potentially contained in GPs 2–5.52

Continued mutations in HP-PRRSVs do not necessarily lead to changes in virus pathogenicity.79 However, some variations in pathogenicity have been observed between HP-PRRSV isolates. In one experimental study, a Vietnamese isolate replicated at lower levels, caused less fever, and resulted in decreased mortality compared to a Chinese HP-PRRSV.53 Natural HP-PRRSV recombinants may exhibit higher or lower pathogenicity compared to their parent strains.52

2. Cleaning and Disinfection

2.1 Survival

2.1.1 PRRSV

PRRSV is inactivated by heat and drying. At temperatures of -70°C to -20°C (-94°F to -4°F), the virus can survive months to years.23 PRRSV is stable at pH 6.5–7.5, but inactivated at high or low pH levels.23 There is evidence that PRRSV can persist in pork. In one study, PRRSV was detected in experimentally contaminated fresh sausage for up to 15 days at 4°C (39.2°F) and for 30 days at -20°C (-4°F).54 However, PRRSV could not be isolated from experimentally contaminated ham, bacon, or acidified sausage. A similar study found that in experimentally contaminated fresh pork, PRRSV could be detected for up to
48h at ambient temperature. At 4°C (39.2°F), PRRSV survived for 3–6 days (depending on the virus concentration used to contaminate the pork). When pork was frozen, PRRSV was detectable for 7–60 days, again varying according to virus concentration. It may be possible for exsanguinated carcasses held at 4°C (39.2°F) to retain infective doses of PRRSV. Despite these findings, the likelihood of PRRSV introduction through the importation of pork is unlikely.

2.1.2 Highly Pathogenic PRRSV
No information is available regarding survival of HP-PRRSV.

2.2 Disinfection
2.2.1 PRRSV
Arteriviruses are reportedly inactivated by lipid solvents including chloroform and ether. Detergent solutions are capable of disrupting the viral envelope. PRRSV is susceptible to phenol, formaldehyde and common disinfectants. Experimentally, room temperature inactivation of PRRSV has been achieved with chlorine (0.03% for 10 minutes), iodine (0.0075% for 1 minute), and a quaternary ammonium compound (0.0063% for 1 minute).

2.2.2 Highly Pathogenic PRRSV
No information is available regarding disinfection of HP-PRRSV.

3. Epidemiology

3.1 Species Affected
3.1.1 PRRSV
Pigs are the natural hosts for PRRSV.

3.1.2 Highly Pathogenic PRRSV
HP-PRRSV infection has been reported only in swine. In addition to domestic pigs, natural infection has been documented in hybrid wild pigs (75% wild boar, 25% domestic Duroc lineage) in China. Wild pigs have been infected with HP-PRRSV experimentally.

3.2 Zoonotic Potential
There is no evidence that PRRSV or HP-PRRSV are zoonotic.

3.3 Geographic Distribution
3.3.1 PRRSV
PRRSV is endemic in many, but not all, swine-producing regions of the world. Countries thought to be PRRS-free include Sweden, Norway, Finland, and Switzerland, as well as New Caledonia, New Zealand, and Australia. In South America, Argentina and Brazil remain PRRS-free, as do Cuba and some areas of the Caribbean.

3.3.2 Highly Pathogenic PRRSV
HP-PRRSV first emerged in China and continues to circulate there. The virus has also spread to Southeast Asia, including Vietnam, the Lao People’s Democratic Republic, Thailand, Cambodia, and India. To date, HP-PRRSV has not been detected in the U.S.

3.4 Morbidity and Mortality
3.4.1 PRRSV
There is little information available on PRRS prevalence, and some estimates may be confounded by the presence of vaccine-derived antibodies. According to the Swine Health Monitoring Project, PRRSV incidence in North America decreased to about 25% in 2013–2014 compared to 35% in previous years.
In many countries, 60–80% of pigs are thought to be infected. Postweaning mortality can be markedly increased in a PRRSV-infected herd. Mortality in sows ranges from 1–4%, with up to 10% reported in a few severe cases. Dead pigs may account for 0–100% of a congenitally infected litter.

3.4.2 Highly Pathogenic PRRSV
HP-PRRSV is known to cause higher morbidity and mortality compared to PRRSV. In the original 2006 Chinese outbreak, morbidity rates of 50–100% were reached. An overall mortality rate of nearly 20% was observed, with up to 100% mortality in individual herds. Mortality rates were highest in suckling pigs (100%), followed by nursery pigs (70%), finishers (20%), and pregnant sows (10%). Similar morbidity and mortality rates have been reproduced experimentally. In wild pigs, experimental infection with HP-PRRSV has resulted in a mortality rate of 25%. In Chinese pigs infected with 2013–2014 HP-PRRSV isolates (recombinants of HP-PRRSV and a recently imported North American virus), morbidity of 100% and mortality of nearly 77% were reported.

4. Transmission
4.1 PRRSV
Infected pigs shed PRRSV in nasal secretions, saliva, urine, semen, mammary secretions, and occasionally feces. PRRSV is transmitted directly via intranasal, oral, intrauterine/transplacental, and vaginal routes. Additionally, intramuscular inoculation has been successfully used to reproduce the disease under experimental conditions. The most effective route of PRRSV transmission seems to be parenteral. Any break in the skin, including those related to husbandry practices, such as ear notching, tail docking, teeth clipping, and injection, can facilitate virus entry. PRRSV can also be spread via bites, cuts, scrapes, or any other activity that involves exchange of blood and oral fluids. PRRSV can be shed in semen for up to 92 DPI, making sexual transmission possible during natural breeding or artificial insemination. Aerosol transmission has also been established. PRRSV has been shown to survive in pork (see section 2 for more information). However, the likelihood of transmission via ingestion of pork and pork products is thought to be quite low, especially if meat is cooked. Indirect transmission occurs following exposure to fomites, including contaminated needles or personnel contaminated with blood or oral fluids, as well as mechanical insect vectors such as houseflies and mosquitoes.

PRRSV endemicity in a herd is driven by prolonged infection in carrier animals (more than 200 days) and the presence of susceptible animals (usually suckling pigs) in a herd. Vertical transmission is a major contributor to within-herd PRRSV transmission (via birth of viremic pigs), as is comingling of infected and susceptible pigs. Between herds, PRRSV transmission occurs mainly following exposure to infected pigs, contaminated semen, or infective aerosols. Proximity to infected herds is a known risk factor for PRRSV transmission.

4.2 Highly Pathogenic PRRSV
It is not known how transmission of HP-PRRSV differs from PRRSV, if at all.

5. Infection in Swine/Pathogenesis
5.1 Pathogenesis
5.1.1 PRRSV
Primary viral replication takes place in macrophages and spreads to lymphoid tissues (spleen, thymus, tonsil, lymph nodes), lungs, and other tissues, where replication continues in monocyte-derived cells. Specific cells known to support PRRSV replication include pulmonary alveolar macrophages (PAMs) and pulmonary intravascular macrophages (PIMs) in the lungs. These cells display the glycoprotein receptor sialoadhesin and the transmembrane protein CD163, which mediate virus entry and release of internalized
virus to the cytoplasm (a prerequisite for viral replication). PRRSV crosses the placenta after day 72 of gestation.

5.1.2 Highly Pathogenic PRRSV
HP-PRRSV has a broader tissue tropism in vivo compared to PRRSV; the immune organs and lungs are severely affected. Viral antigen has been detected in the trachea, esophagus, liver, mandibular gland, and thyroid gland in pigs inoculated with HP-PRRSV but not PRRSV. Similarly, epithelium in tissues including the interlobular bile duct in liver, distal renal tubule of kidney, esophageal gland and tracheal gland have been found to be positive for viral antigen only in HP-PRRSV-inoculated pigs. HP-PRRSV is also known to cross the blood-brain barrier and induce damage to neurons and neuroglial cells. In an experimental study comparing the pathogenicity of HP-PRRSV in wild and domestic pigs, HP-PRRSV-positive cells were found in the bronchiolar, gastric, and renal tubular epithelial cells in wild pigs but not in domestic pigs.

5.2 Clinical Signs
5.2.1 PRRSV
Clinical presentation is variable and influenced by a number of factors including virulence of the infecting strain, host immune status and susceptibility, exposure to lipopolysaccharides (bacterial endotoxin), presence of concurrent disease (see section 6.1 for more information), and management factors. In breeding animals, anorexia, fever, lethargy, depression, respiratory distress, and vomiting can be seen, as well as cyanosis of the ears, abdomen, and vulva. Transplacental transmission results in reproductive failure (stillborn, autolyzed, and/or mummified fetuses) or birth of viremic piglets. In young, growing, and finishing pigs, acute viremia leads to respiratory disease. In addition to pneumonia, sneezing and expiratory dyspnea, fever, lethargy, and depression can be seen. The peak age for respiratory disease is 4–10 weeks. Where PRRSV is endemic, disease is seen mostly in nursery pigs (when maternal antibody wanes) or in replacement gilts or sows. Epidemics occur when the virus enters an immunologically naïve herd or when antigenic variation leads to the emergence of a new PRRSV variant.

5.2.2 Highly Pathogenic PRRSV
The first HP-PRRSV epidemic, described in China in 2006, was characterized by fever up to 40–42°C (104–107.6°F), neurological signs such as shivering, petechiae, an erythematous blanching rash, and blue ears. Lameness, respiratory signs, and diarrhea were also reported, as well as depression, anorexia, and lethargy. Similar clinical signs have been described in HP-PRRSV outbreaks occurring since 2006. Signs consistent with the original outbreak have also been reproduced in pigs experimentally infected with HP-PRRSV. Unlike PRRSV, HP-PRRSV causes clinical disease and death in adult pigs, including pregnant sows.

5.3 Postmortem Lesions
5.3.1 PRRSV
From 4 to ≥28 DPI, gross and microscopic lesions are observed in the lungs and lymph nodes, where viral replication predominantly occurs. Interstitial pneumonia, which varies from multifocal to lobular to diffuse in distribution, is most severe from 10–14 DPI and lungs may appear mottled and tan. Lung lesions may be complicated by concurrent bacterial and/or viral infections. Enlarged, tan, edematous lymph nodes are also commonly seen. Endometritis, myometritis, and placental lesions may occur in infected sows; in aborted fetuses and stillborn pigs with uncomplicated PRRS, no lesions may be detectable, but umbilical cord arteritis and hemorrhage may be seen, as well as lung and lymph node lesions similar to those seen in older pigs. Microscopic lesions associated with PRRSV include nonsuppurative interstitial pneumonia, mild nonsuppurative encephalitis, myocarditis, rhinitis, and possible depletion of germinal centers of lymph nodes. Although these lesions may be suggestive of PRRSV they are not pathognomonic.
5.3.2 Highly Pathogenic PRRSV
In the 2006 Chinese outbreak, HP-PRRSV infection led to severe, multiorgan damage. Lesions described by Tian et al. include: foci with pathological changes and hyperplasia in the lungs, with lung hemorrhagic spots and lung edema; spleen infarcts and bladder dilatation, filled with reddish brown urine, and frequent blood spots in the kidney; putrescence of cardiac muscle; foci of yellow-white necrosis or hemorrhage in the liver; slightly softened encephala, blood egression emission, and effusion of jelly from the brain putamen; obvious hemorrhagic spots in the lymph nodes; arthritis with swollen joints; and intestinal ulceration.

Pigs experimentally infected with HP-PRRSV have demonstrated acute lung injury including distortion of lung structure and diffuse fibrosis. Severe histopathological lesions have been described including destruction of lung structure with extensive hemorrhage and a large number of inflammatory cells infiltration, severe dropout of alveolar epithelial cells with exudation of inflammatory cells and erythrocytes into the alveolar spaces, vasculitis and thrombus, alveolar lumens flooded with edema fluid and pulmonary interstitial edema accompanied vascular trauma, and formation of multinucleated giant cells and epithelioid cells. Lung lesion scores in experimentally infected domestic pigs are significantly higher than in wild pigs.

6. Diagnosis
6.1 Clinical History
PRRS may be suspected when reproductive disease occurs in breeding swine and respiratory disease is present in pigs of any age. However, presentation is variable and may be complicated and/or compounded by other pathogens including swine influenza virus, Streptococcus suis, Mycoplasma hyopneumoniae, Salmonella choleraesuis, Haemophilus parasuis, Pasteurella multocida, porcine circovirus, porcine respiratory coronavirus, and Actinobacillus pleuropneumoniae. PRRSV is the most common virus isolated in cases of porcine respiratory disease complex (PRDC). Other differential diagnoses for respiratory and postweaning disease include hemagglutinating encephalomyelitis virus, syncytial pneumonia and myocarditis, post-weaning multisystemic wasting syndrome and Nipah virus. Differential diagnoses for reproductive disease include classical swine fever, African swine fever, leptospirosis, porcine parvovirus, porcine enterovirus, hemagglutinating encephalomyelitis virus, Toxoplasma gondii, and Aujeszky’s disease (pseudorabies virus).

6.2 Tests to Detect Nucleic Acids, Virus, or Antigens
6.2.1 PRRSV
Virus isolation can be done in PAMs or African monkey kidney cells (line MA-104, sublines CL-2621 or MARC-145). The optimal time frame for virus isolation from serum and tissues is 1–35 DPI, although virus may persist in the lymphoid tissues. Following isolation, viral antigen can be visualized via direct fluorescent antibody (using fresh infected tissues, lung preferred) or immunohistochemistry (IHC) on formalin fixed tonsil or lung. Viral RNA can be detected by reverse transcriptase polymerase chain reaction (RT-PCR) or in situ hybridization. A one-step SYBR Green-based RT-PCR method to detect PRRSV has been developed though this test is mostly obsolete in the U.S. Immunochromatographic strip tests for PRRSV detection have been described, and loop-mediated isothermal amplification (LAMP) assays targeting both type 1 and type 2 PRRSV have been developed for experimental use. These tests are not widely available or routinely used in the U.S.

6.2.2 Highly Pathogenic PRRSV
Traditional RT-PCR PRRSV assays cannot discriminate between PRRSV and HP-PRRSV. However, newer RT-PCR variations available in China are capable of differentiation. These tests are not yet widely available in the U.S. Assays described experimentally that are capable of differentiation include a
multiplex quantitative RT-PCR (qRT-PCR) assay, as well as a one-step RT-PCR assay and a nested RT-PCR assay that detect changes in NSP2.

A SYBR Green-based qRT-PCR assay and a duplex qRT-PCR assay capable of differentiating HP-PRRSV from type 2 PRRSV have been developed for experimental use. An immunochromatographic strip test described experimentally can also detect both PRRSV and HP-PRRSV. A novel fluorescent probe-based real-time reverse transcription recombinase polymerase amplification (real-time RT-RPA) assay that detects HP-PRRSV has recently been described.

Presumably, ORF5 or complete genome sequencing followed by phylogenetic analysis would also lead to the accurate identification of HP-PRRSV.

6.3 Tests to Detect Antibody

6.3.1 PRRSV
Serology is useful to confirm the presence of PRRSV in a herd. Single serum samples have limited diagnostic value since PRRSV is ubiquitous and results may be confounded by previous vaccination and/or the presence of maternal antibody. To diagnose active infection, seroconversion must be demonstrated by testing acute and convalescent samples.

Indirect fluorescent antibody (IFA) is capable of detecting both IgM (as early as 5 DPI) and IgG (as early as 9–14 DPI). This test is used mostly to confirm suspected false-positive enzyme-linked immunosorbent assay (ELISA) results. A commercial ELISA (IDEXX PRRS X3 ELISA, IDEXX Laboratories Inc.) targeting the nucleocapsid protein is the reference standard. This test is capable of identifying type 1 and type 2 PRRSV. IgM antibodies are detectable as early as 9 DPI; they peak at 30–50 DPI and then decline over 4–12 months to negative levels. Prolonged carrier pigs may test negative via ELISA. An ELISA using a recombinant NSP7 as antigen has been tested experimentally. The virus neutralization (VN) assay is highly specific but not a routine diagnostic test.

6.3.2 Highly Pathogenic PRRSV
No information was found on the identification of antibodies specific to HP-PRRSV. ELISAs developed for PRRSVs (e.g., IDEXX PRRS X3 ELISA, IDEXX Laboratories Inc.) may also detect antibodies to HP-PRRSV.

6.4 Samples

6.4.1 Preferred Samples

6.4.1.1 PRRSV
Samples suitable for virus detection include serum, lung, lung lavage, lymph nodes, spleen, heart, kidney, thymus, and tonsil (especially in chronically infected pigs). PRRSV can also be detected in oral fluids. Tissues should be collected from weak-born neonates that have not nursed, nursing pigs that are clinically ill, or febrile, anorectic postweaned pigs or sows. Virus can also be detected in semen by RT-PCR. Tissues or fluids from aborted, mummified, or stillborn pigs are generally of limited diagnostic value; however, they are sometimes used for RT-PCR or ruling out other infections.

6.4.1.2 Highly Pathogenic PRRSV
Tissue samples appropriate for detection of PRRSV are also suitable for HP-PRRSV.

6.4.2 Oral Fluids
Experimentally, PRRSV can be detected in oral fluids by qRT-PCR and antibody can be detected by ELISA and IFA. A commercial ELISA to detect PRRSV in oral fluids is available (IDEXX PRRS Oral Fluids Ab Test, IDEXX Laboratories, Inc.). Pen-based oral fluid sampling may be useful for PRRSV surveillance in swine production systems.
7. Immunity

7.1 Post-exposure

7.1.1 PRRSV

PRRSV inhibits type I IFNs and suppresses innate immunity in infected pigs.\(^9^2\) Cell-mediated immune responses peak around 10 weeks PI.\(^2^2\) Neutralizing antibodies against the viral proteins N and GP5, and NSPs 1, 2, and 7 appear about 4 weeks PI.\(^2^2\) Although PRRSV can persist long-term in carrier pigs, it seems that most infected pigs become immune and viral shedding usually stops sometime before 60 DPI. Declining antibody titers have been observed around 4–8 months PI.\(^5^8\) In piglets, passive immunity declines soon after weaning. Adult animals are much more resistant to PRRSV infection compared to weaned pigs.\(^2^3\)

7.1.2 Highly Pathogenic PRRSV

HP-PRRSV seems to induce higher levels of anti-PRRSV IgG antibody in wild pigs compared to domestic pigs at 21 DPI.\(^6^1\) Experimentally, HP-PRRSV is also a stronger inducer of toll-like receptor (TLR) 3, 7, 8 expression and IL-1β, IL-6, TNF-α, IFN-γ production compared to PRRSV.\(^7^2\)

7.2 Vaccines

7.2.1 PRRSV

Vaccination against PRRSV may result in protective immunity, moderate clinical signs, and reduce shedding of virus\(^2^3\); however, experience in the field suggests that vaccination is inconsistently effective.\(^2^3\) Inactivated and modified live vaccine (MLV) formulations are available. Inactivated vaccines are most effective when used in combination with MLVs or in previously infected animals. MLVs induce a more efficacious immune response compared to inactivated vaccines, though these formulations have been known to revert to virulence under field conditions.\(^2^3\) MLVs provide homologous protection but due to the genetic diversity of PRRSV, cross-protection between vaccine and field strains may not be sufficient to prevent infection.

Development of a universal vaccine has been suggested as a way to overcome current vaccine limitations.\(^9^3\) Other vaccine technologies being explored include recombinant vectors (e.g., recombinant adenovirus, attenuated pseudorabies virus, recombinant transmissible gastroenteritis virus, \textit{Mycoplasma} spp.), marker vaccines (differentiating infected from vaccinated animals [DIVA] and compliance marker vaccines), infectious PRRSV cDNA clones, use of as PRRSV as a viral vector (for expression of marker genes, pathogen genes, and cytokine genes), chimeric PRRSVs, PRRSV attenuation through codon pairs deoptimization of the major envelope gene, and DNA shuffling (rapidly accelerated mimicry of natural recombination).\(^9^4\)

7.2.2 Highly Pathogenic PRRSV

As with PRRS vaccines, attenuated HP-PRRSV vaccine strains such as JXA1-R, HuN4-F112, and TJM-92 have demonstrated protection against homologous challenge.\(^9^5\)-\(^9^8\) Partial heterologous protection has also been reported following vaccination with attenuated type 1\(^9^9\) and type 2\(^1^0^0\)-\(^1^0^3\) MLVs. The HP-PRRSV vaccine strain JXA1-R has been shown to provide heterologous protection against NADC-20.\(^1^0^4\) New vaccine candidates are continuing to be described. Current candidates include a highly attenuated derivative from the HP-PRRSV strain QY1, showing a 32-aa deletion in NSP2\(^1^0^5\) and the HP-PRRSV strain HB-XL, suspected to be a novel virus caused by vaccine recombination that results in low morbidity and mortality in pigs.\(^4^0\)

Safety must be taken into account in HP-PRRSV vaccination programs. In China, at least two instances of natural recombination between a PRRSV vaccine strain and a recently circulating HP-PRRSV have been documented.\(^1^0^6,1^0^7\) HP-PRRSV revertants of a vaccine strain (JXA1-P80) have also been detected in Chinese pigs.\(^1^0^8\)
7.3 Cross-protection
As previously noted, PRRSV evolves rapidly, resulting a genetically and antigenically diverse population. Recombination is also a driver of virus change, and it has been linked to the emergence of novel HP-PRRSV strains in China in recent years. Infection with one PRRSV strain does not prevent infection with another. Herds can be simultaneously infected with more than one PRRSV.

8. Prevention and Control
There is no treatment for PRRS. Antibiotics are often used to combat secondary bacterial infections. Methods to prevent the entry of PRRSV into a naïve swine herd include quarantine and testing of new breeding animals; sanitation and drying of transport/supply vehicles; shower-in, shower-out or use of Danish entry systems; and implementation of insect control program (e.g., screening, habitat management, and insecticide use). Air filtration systems have been shown to reduce the risk of PRRSV transmission on sow farms, boar studs, and nurseries, though initial investment can be costly.

In PRRS-endemic herds, control programs can be implemented to limit adverse effects of the disease. Best control strategies will vary depending on specific farm situations. PRRS control programs often involve a combination of the following:

- Gilt acclimatization—use of replacement gilts that have developed PRRSV immunity prior to herd introduction through vaccination, contact with PRRSV-infected animals, or intentional exposure to PRRSV (e.g., feedback of PRRSV-contaminated tissues or exposure to serum from acutely infected pigs);
- Breeding herd control—use of acclimatization protocols for all incoming breeding stock as described above, use of PRRSV-negative semen, limited new stock introductions, and consideration of herd closure for at least 200 days;
- Pig management—restricted cross-fostering, euthanasia of affected pigs, and use of all-in, all-out pig flow in the nursery. Partial depopulation can prevent lateral spread of PRRSV, but in herds where that is not possible, mass MLV vaccination can also be employed;
- Eradication—elimination of PRRSV from a herd via total herd depopulation/repopulation, partial depopulation, segregated early weaning, test and removal, and herd closure.

In China, HP-PRRSV control programs have mostly centered on vaccination, though a commonly used product (Ingelvac PRRS MLV, Boehringer Ingelheim) provides only partial protection. Acclimatization of pigs (through exposure to serum from PRRS-positive pigs) has also been done, but whole herd depopulation/repopulation, test and removal, and herd closure strategies have not been implemented widely in China.

PRRSV is an OIE-listed disease. However, it is not covered in the 2016 OIE Terrestrial Animal Health Code.

10. Gaps in Preparedness
PRRSVs have been economically devastating for swine producers in many parts of the world including the U.S., and new PRRSV variants—causing even higher morbidity and mortality rates—continue to emerge in Asia. Preventing the spread of PRRSV within and between farms is critical to reduce the burden of disease in China and other countries affected by HP-PRRSV. North American PRRSVs have previously been imported into China. The scenario should be considered in which Chinese HP-PRRSVs are imported into the U.S. Potential entry routes into the U.S. should be investigated. Strict biosecurity measure should be in place to prevent an HP-PRRSV incursion.
Although PRRSV was identified more than 25 years ago, many knowledge gaps remain. To advance our understanding of PRRSV, future studies on basic viral biology, pathogenesis, host genomics, viral genomics, and immunology must continue. Although PRRSV vaccines are available, control programs are hampered by the virus’ rapid evolution and deficient heterologous coverage. New vaccine technologies must be explored to enhance cross-protective immunity and produce products that cannot revert to virulence. Prolonged carriage in pigs impedes elimination efforts, and diagnostic tests that can differentiate infected from vaccinated animals (DIVA) are urgently needed. Improved vaccines and diagnostics are critical components in a PRRSV control or elimination program. To date, pork industry funded research has focused on immunology and vaccine development; epidemiology, risk factors, and control strategies; diagnostic tests and PRRS surveillance; regional elimination; and genetic resistance to disease.
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