Chapter 1

Introduction

Porcine reproductive and respiratory syndrome (PRRS) is one of several major swine diseases causing economic loss in the swine industry worldwide including Thailand. Clinically, PRRS is characterized by reproductive failure in gilts and sows, and respiratory diseases in young pigs. Morever, PRRS virus (PRRSV) increased susceptibility to other respiratory bacterial infection (Thanawongnuwech et al., 2000; Wills et al 2000). In Thailand, serological studies suggest the widespread of PRRSV infection among herds and the earliest detection of seropositive animals was found in 1989 (Damrongwatanapokin et al., 1996). The virus was first successfully isolated from suckling and nursery piglets with severe chronic respiratory distress in June 1995. By an indirect immunofluorescent staining and reverse-transcription polymerase chain reaction (RT-PCR) indicated that the Thai isolate was more closely related to the American strain than the Lelystad virus (Damrongwatanapokin et al., 1996). The nested multiplex PCR demonstrated that both EU and US genotypes have been present in Thailand since Thailand has continuously imported swine breeders from both European and North American countries (Thanawongnuwech et al., 2002).

In order to control PRRSV, a commercial modified live virus (MLV) vaccine has been used in the United States since late 1994 and has had some limited use in the other countries, including Thailand. Although the vaccine virus is attenuated, it

occasionally persist for at least several weeks in a vaccinated pig (Wesley et al., 1998a). Furthermore, the live attenuated vaccine virus may spread to non - vaccinated sows, change genetically or revert to virulence under the field conditions (Nielsen et al., 2001) such as the incidence of the presence of vaccine virus in pigs in Denmark (Madsen et al., 1998), in Korea (Cheon and Chae, 2000), in Japan (Itou et al., 2001) and in Canada (Cai et al., 2002). It is hard to distinguish a particular vaccine viruses from PRRSV field strains by the routine laboratory procedures. Recently, a Polymerase Chain Reaction – based Restriction Fragment Length Polymorphism (PCR–based RFLP) analytic method has been developed for helping grouping the PRRSV (Wesley et al., 1998a). The selected restriction enzyme cut at the precise 4-6 position of DNA sequence. Therefore, the mutation of the sequence could change the cutting site of DNA resulting in the different RFLP patterns. The various patterns are used for grouping the viruses. The grouping using RFLP patterns will be a valuable tool in farm management and epidemiologic studies. The RFLP may indicate the mutation of viral or may differentiate the infected pig from the vaccinated pig (Wesley et al., 1998a). However, the MLV vaccine is not officially allow for use in Thailand.

The hypothesis of this study was to prove the genetic variation and the differences of virulence among the selected Thai isolates using RFLP analysis and the challenge model in PRRSV- free pigs.

The objectives of this study were to study the RFLP patterns of the Thai isolates and to study the pathologenesis of the Thai isolates of each genotype. This thesis is

divided into two parts. The first part consists of the PCR - based RFLP of the open reading frame 5 (ORF5) analysis using for grouping the Thai isolates of PRRSV. The ORF5 is the hypervariable region containing the variation of the restriction pattern. The second part is a challenged model studying the pathogenesis of PRRSV in Thailand. The virus isolation, antibody detection (enzyme - linked immunoabsorbent assay), antigen detection (immunohistochemistry) and viral nucleic acid detection (multiplex polymerase chain reaction) were used to determine the presence of PRRSV infection. Pathological study including gross and microscopic findings were demonstrated for the virulence of the selected Thai isolates. The results from this experiment could be useful for diagnosis and pathogenesis of PRRSV infection in Thailand.