5. DISCUSSION AND CONCLUSIONS

5.1 Discussion

The whole study on a pork chain was divided into two parts, breeder and fattening farms in the region of Chiang Mai province, Thailand. The breeder farm was an integrated pork production company and the fattening farms were contract farms. After weaning at day 18-21 days, piglets were transported to fattening farms and fattened up to slaughter at the age of 180 days and a weight of 90-100 kg. The breeder farm and subsequently the fattening farms were forthcoming in supplying any information needed. The company's good information system provided a food trail and responsibility for consumers' safety.

5.1.1. Materials and Methods

The amount of each faecal sample was 25 g which was sufficient for investigation according to ISO 6579.

Most of studies investigating the prevalence of *Salmonella* in pig feces use reference culture methods. Samples from herds with peak clinical salmonellosis can be easily identified directly and without any enrichment by plating on selective agars, whereas samples from chronically infected pigs or from the environment always require pre-enrichments and selective enrichments.

In this study, tetrationate broth and modified semisolid Rappaport-Vassiliadis medium were used as selective broth media. BPLS and XLT4 agar were used as solid selective enrichment. The characteristic of XLT4 is a high degree toward inhibition of

other competing bacteria. Significantly increasing in the recovery of *Salmonellae* and essentially eliminating false positive suspected colonies are consequences of using XLT4.

Modified semisolid Rappaport-Vassiliadis medium (MSRV) could be more appropriate than liquid Rappaport-Vassiliadis medium (RV) for the examination of swine feces. In 2001, Voogt *et al*. found that the performance for *Salmonella* detection of MSRV was significantly better than RV. However, the large microbial load of feces, with highly competing background floras, can hamper more; MSRV is intended for the detection of motile *Salmonellae* and is less appropriate for the detection of nonmotile *Salmonellae* (Malorrny and Hoorfar, 2005).

5.1.2. Results

Results of investigations into animal samples (faecal swab, 25 g of feces and skin swab) provided an estimate of the prevalence of current *Salmonella* infections in piglets and in the fattening period at different times (7, 18, 60, 90 and 120 days). The prevalence of faecal swab sample, 25 g of feces and skin swab sample was 4.74%, 23.08% and 1.93% respectively. The number of *Salmonella* isolates increased from day 18, 60, 90 and 120 (5%, 11.36%, 11.36% and 34.88%). Overall prevalence of *Salmonella* in piglets (per sample) was 12.32% (26/211).

For environmental samples (feed, floor swab and water), the prevalence of feed, floor swab and water was 10%, 20% and 10% respectively. These results indicate the level of *Salmonella* contamination in the environment or *Salmonella*.

The incidence of *Salmonella* increased during day 7-18, 18-60, 60-90 and 90-120 (2.5, 7.41, 8.11 and 57.14). The highest incidence of *Salmonella* was during day 90-120.

Comparison of *Salmonella* prevalence in the pork chain project

	Sow Ngasaman, 2007	Piglet This study	Finishing pig Dorn-in, 2005
%prevalence	20%	12.32%	62.9%

The present study shows that *Salmonella* prevalence in pre-weaning piglets is lower than post-weaning piglets. The explanation is that maternal antibodies may passively protect piglets during the first weeks but these antibodies decrease after a few weeks. When these maternally derived antibodies decrease, piglets are no longer protected and environmental *Salmonella* may contaminate them. Proux *et al*. (2000) reported that maternal antibodies persisted until 7 weeks of age and post-*Salmonella* contamination seroconversion was detected from the 8th week of age onwards. Furthermore, Funk *et al*. (2001) explained the spectrum of maternal antibodies secreted in the colostrum (I_gG) and milk (I_gA) might differentially protect the piglets against colonization of different *S*.enterica serotypes.

The prevalence of piglets shedding *Salmonella* before weaning was relatively low. However the prevalence was likely underestimated as a consequence of the low sensitivity of rectal swabs for detection of *Salmonella* from feces (Funk *et al.,* 2000).

Weaning stress and horizontal transmission occurred in the nursery were the important factors of increasing the prevalence in the nursery. The most important stress factors are likely to be i) a change in feed, ii) the commingling of litter, and iii) a piglet's being deprived of the antibodies found in sow's milk before the activation of its own immune response (Kranker *et al.*, 2003).

5.1.3. Serotypes of Isolates

From 33 positive samples, two serotypes were identified, *S*.Typhimurium and *S*.Rissen. The proportion of serotypes identified in the pork chain project are summarized in the table below.

Comparison of *Salmonella* prevalence and serotype in the pork chain project

Serotype	Sow Ngasaman, 2007	Piglet This study	Finishing pig Dorn-in, 2005
S.Typhinmurium	-	6.06%	18.3%
S.Rissen	50%	18.18%	45.4%
S.Stanley	25%	-	11.5%
F-67	-	75.76%	NA

NA = Not available

For comparison purposes, the top five of the most frequent *Salmonella* serotypes in human cases in Thailand were *S*.Enteritidis (18.46%), *S*.Stanley (10.68%), *S*.Choleraesuis (7.95%), *S*.Rissen (7.64%) and *S*.Weltevreden (6.89%) (NSSC, 2006). *S*.Typhimurium ranked 9th in this investigation.

During the last few years, in Thailand *S*.Rissen is increasingly isolated from foodborne gastrointestinal infections in humans (1.6% in 1993, 8.2% in 2002, 8.91% in 2005 and 7.64% in 2006) (Bangtrakulnonth *et a*l., 2004; NSSC, 2005; NSSC, 2006). From raw material foods, ready-to-eat foods and animals, *S*.Rissen was isolated in amounts of 21.35%, 13.29% and 25.34% respectively (NSSC, 2005). The results from this study indicate that fattening pigs and the environment in pig fattening farms are an important reservoir for *S*.Rissen.

Padungtod and Kaneene (2006) reported the serotypes and the relationship between pigs and farm workers in northern Thailand. The most common serotype found in pig and farm workers from pig farm was *S*.Rissen. *S*.Rissen was relatively common in farm workers with livestock exposure, which may indicate a common source of this serotype.

S.Typhimurium is a virulent serotype. In Thailand, *S*.Typhimurium isolation from human from 1993, 2002, 2005 and 2006 was 4.7%, 4.1%, 2.64% and 3.28% respectively (Bangtrakulnonth *et al*., 2004, NSSC, 2005 and NSSC, 2006). These

results show that human cases related to *S*.Typhimurium had not increased. The results of this study demonstrate that *S*.Typhimurium exists in pig farms and in farm's environment and subsequently pigs could be an important reservoir for *Salmonella* contamination of the pork chain.

5.1.4. Correlation between *Salmonella* isolation from faecal swabs and 25 grams of Feces

A total of 70 samples was examined both in terms of faecal swab and 25 g of feces. *Salmonella* prevalence in faecal swabs and 25 g of feces in total shows, that the correlation was very low (Kappa= 0.017).

In 2001, Hurds *et al*. reported that the sensitivity of bacteriological culture might vary between 10% and 80% depending on the sampling and testing protocols used. Different fecal sample size (rectal swab, 1, 10 and 25 g) was compared in another study, and an increased sensitivity was associated with increased fecal sample size (Funk *et al*., 2000). In order to develop methods appropriate for Salmonella detection, the nature of organism, cost-effectiveness and the reliability of the tests need to be considered.

Farm management practice is also one of the factors associated with the *Salmonella* prevalence. For pre-harvest control (at farm level), hygiene and husbandry (all-in/all-out system, cleaning and disinfection and biosecurity system) are considerably important. In 1997, Dahl *et al*. found that *Salmonella* infection can be controlled if pigs are moved from the herd to a clean, uninfected and fed only *Salmonella*-controlled feed, which is in contradictive to the results of Funk *et al*. (2001), who found that cleaning and disinfection practices in multi-site swine production system in USA do not reliably eliminate *S. enterica*.

Feed stuff plays an important role in food-chain hygiene, the surveillance for *Salmonella* contamination in animal feed should be conducted. In Thailand, the Department of Livestock (DLD), Ministry of Agriculture and Cooperatives runs a

routine survey as a part of animal-feed quality-control program. In 2006, *S*.Lamphun was discovered, for the first time from animal feed, by the DLD and Cooperatives, in co-operation with the central reference laboratory of the Department of Medical Science (Kusum *et al*., 2006).

Factors that may affect prevalence include those affecting the detection of the organism, such as sampling procedures, bacterial isolation and identification methods; factors that directly influence prevalence, are animal management, slaughter practices and cross-contamination (Bryan and Doyle, 1995).

Finally, results from this investigation into *Salmonella* can not be generalized for pigs raised by other companies in Chiang Mai or even other part of Thailand because this study obtained samples from 1 particular breeder farm and 2 fattening farms.

5.2 Conclusion

The long-term goal of pre-harvest investigations of Salmonella in swine is to determine control measures that will reduce the risk of human foodborne disease. To design effective pre-harvest control programs, an understanding of transmission of Salmonella on farms is essential. Data from this study indicate that variability in *Salmonella* prevalence and serotypes within farm can be substantial. Ability to classify herds by *Salmonella* risk will be necessary to implement the monitoring and control program.

In this study, *Salmonella* prevalence in piglets was low and increased during the fattening period. 2 serotypes (*S*.Rissen and *S*.Typhimurium) were isolated in this investigation. The correlation between investigation results of faecal swab and 25 g of feces was poor.

The selection of a diagnostic test depends not only on test characteristics, but also the purpose of the test (screening of populations, evaluation of the efficiency of interventions, trace back of food-borne outbreaks, monitoring process control etc.). A

clear definition of the purposes of a *Salmonella* diagnostic test must be established prior to initiating any investigations into *Salmonella*.

Further evaluation of the impact of *Salmonella* serovar present on farms with seroprevalence and the relationship of on-farm seroprevalence with food safety risk are needed for pre-harvest *Salmonella* diagnostics in Thailand.