APPENDICES

APPENDIX A

LIST OF CHEMICALS AND REAGENTS USED FOR SALMONELLA CULTURE IN THIS STUDY

1. Culture Media

- Buffered Peptone water (BPW)
- Rappaport Vasiliadis broth (RV)
- Tetrationate (TT broth)
- Brilliant Green Phenol Red Lactose Sucrose agar (BPLS)
- Xylose-lysine-tergitol 4 agar (XLT4)
- Nutrient agar (NA)

2. Biochemical Media

- Triple Sugar Iron agar slants (TSI)
- Urea slant
- Voges-Proskaur broth (VP)
- Motile-indole-lysine broth (MIL)

3. Media for Serogroup Typing and Flagella Phase

- Nutrient agar (NA) + 65% Nutrient agar (NA)

4. Media for Storage of Salmonella Isolates

- 50% Nutrient agar (Half-NA)

5. Other Chemicals and Reagents

- Kovac's reagent
- creatine solution
- ethanolic solution of 1-naphthol
- Potassium hydroxide solution

APPENDIX B

MEDIA PREPARATION

Buffered peptone water (BPW; Merck KGaA, Germany)

Preparation: Suspend **25.5** g in 1 liter of demineralized water; if desired dispense into smaller vessels; autoclave (**15 min. at 121** °C).

pH: 7.0 ± 0.2 at 25 °C

Rappaport Visiliadis broth (RV; Merck KGaA, Germany)

Preparation: Suspend **42.5 g in 1 liter** of demin. water; dispense into tubes or flasks; autoclave (15 min. at 121 °C).

pH: 5.2 ± 0.2 at 25 °C

Tetrathionate broth (TT; Merck KGaA, Germany)

Preparation: Suspend **82 g in 1 liter** of demineralrized water, heat briefly to boiling. **Do not autoclave!** After cooling, add 20 ml/l iodine potassium iodine solution and 10-ml/l 0.1% brilliant green solution (brilliant green, Cat. No.1.01310.). Dispense any eventual precipitate.

Brilliant Green Phenol Red Lactose Sucrose Agar (BPLS; Merck KGaA, Germany)

Preparation: Suspend **51 g in 1 liter** of demin. water by heating in a boiling water bath or in a current of stream; autoclave (15 min. at 121 °C); pour to plates.

pH: 6.9 ± 0.2 at 25 °C

Xylose-lysine-tergitol 4 (XLT4; Merck KGaA, Germany)

Preparation: Suspend **59 g in 1 liter** of demin. water, add 4.6 ml XLT4 Agar Supplement solution and heat the medium in a boiling water-batch (*not* on a heating-plate!).Cool to approx. 50 °C and pour plates. **Do not overheat, do not autoclave!**

pH: 7.4 ± 0.2 at 25 °C

Triple Sugar Iron Agar Slants (TSI; Merck KGaA, Germany)

Preparation: Suspend **65 g in 1 liter** of demin. water by heating in a boiling water bath or in a current of stream; dispense into tubes; autoclave (15 min. at 121 °C). Allow solidifying to give agar slants.

pH: 7.4 ± 0.2 at 25 °C

Urea Agar (Urea; Merck KGaA, Germany)

- 1. Suspend 29 g of medium in 100 ml of demineralized water.
- 2. Mix thoroughly and sterilize by filtration.
- 3. Dissolve 15 g of agar in 900 ml of demineralized water.
- 4. Sterilize by autoclaving at 121°C for 15 minutes.
- 5. Cool to 45-50°C and aseptically add the sterile Urea Agar Base.
- 6. Mix thoroughly and dispense into sterile tubes.
- 7. Cool in a slanted position so that deep butts are formed.

Nutrient Agar (NA; Merck KGaA, Germany)

Preparation: Suspend **20 g in 1 liter** of demineralized water by heating in a boiling water bath or in a current of stream; autoclave (15 min, at 121 °C). Pour to plates.

pH: 7.0 ± 0.2 at 25 °C

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APPENDIX C

HISTORY OF SUBJECTS (THE SLAUGHTER PIGS)

Table 1: Sources (districts and provinces) of slaughter pigs (LP = Lumphun Province, CM = Chiang Mai Province)

Slaughterhouse visit	Date collected	Numbers of studied pigs	Location of farms of origin	Farm type (closed/open)
1 st	Dec.20, 2004	8	Mae-tha (LP)	Open
2 nd	Jan.7, 2005	10	Ban-thi (LP)	Open
3 rd	Jan.14, 2005	10	San-sai (CM)	Closed
4 th	Jan.25, 2005	10	San-sai (CM)	Open
5 th	Feb.4, 2005	10	San-sai (CM)	Open
6 th	Mar.4, 2005	8	Pa-sang (LP)	Open
7 th	Mar.8, 2005	8	Mae-tang (CM)	Open
8 th	Mar.10, 2005	8	Mae-rim (CM)	Open
9 th	Mar.16, 2005	8	Pa-sang (LP)	Open
10 th	Mar.18, 2005	8	Mae-tang (CM)	Open
11 th	Mar.25, 2005	7	Mae-wang (CM)	Open
12 th	Mar.29, 2005	10	San-kam-phang (CM)	Closed
13 th	Apr.2, 2005	8	Mae-tha (LP)	Open
14 th	Apr.5, 2005	10	San-kam-phang(CM)	Closed
15 th	Apr.20, 2005	10	San-kam-phang (CM)	Closed
16 th	Apr.24, 2005	8	Mae-on (CM)	Open
17 th	Apr.27, 2005	8	San-kam-phang(CM)	Open
18 th	Apr.30, 2005	118812	Mae-taeng (CM)	Open //
19 th	May4, 2005	8	Mae-tha (LP)	Open
20 th	May6, 2005	8	Mae-tha (LP)	Open
21 st	May9, 2005	8	Mae-tha (LP)	Open
Total	21 visits	181 pigs	2 provinces	2 types

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DECLARATION

I, the undersigned, declare that the thesis is my original work and has not been presented for a degree in any University.

Name:Signature:	Wasan Chantong Wasan Chamlong		
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