APPENDICES

APPENDIX A

EQUIPMENT, MATERIALS, MEDIA AND REAGENTS

1. Lab Equipments and Materials

- Sterile 500, 1000 and 2000 ml Erlenmeyer flasks, sterile 250 and 500 ml beakers, and containers of capacity to accommodate samples
- Balance with a 2000 g-weights capacity and a sensitivity of 0.1 g
- Incubator, 37 and 42 C
- Laboratory refrigerator, - 20 C and -1 to 4+ C
- Water bath
- Sterile spoons for transferring faecal samples and media
- Sterile culture dishes, 15*100 mm, glass or plastic
- Sterile pipettes
- Inoculating needle and inoculating loop (10 micrometer)
- Culture tubes, 16*150 and 20*150 m
- Test or culture tube racks
- Vortex mixer
- Stomacher machine.
- Sterile scissors, scalpel, and forceps
- Bunsen burner
- Stomacher bags and plastic bags
- Appendop
- Autoclave

2. Equipment and Material for Sample Collection

- Sterile cotton sock swabs
- Disposable hand gloves
- Stomacher bags and plastic bags
- Buffered peptone water (BPW)
- Sterile 1000 ml. Duran bottle
- Marker pens
- Alcohol, cotton, lighter
- Normal saline
- Disposal gloves, boots and lab coat
- Ice box with ice
- Snare

3. Media, Reagent and Chemicals

- Buffered Peptone Water (BPW)
- Nutrient agar (NA)
- Brilliant-green Phenol-red Lactose Sucrose Agar (BPLS)
- Xylose Lysine Tergitol 4 agar (XLT4)
- Muller Kaufmann Tetrathionate broth (MKTT)
- Rppaport-Vassiliadis broth (RV)
- Triple Sugar Iron Agar (TSI)
- Urea Agar
- Motility Indole Lysine Decarboxylation (MIL)
- Voges-roskauer Reaction (VPR)
- *Salmonella* polyvalent somatic (O) antiserum A- E
- *Salmonella* polyvalent somatic (O) antiserum F- 67
- *Salmonella* somatic (O) antiserum- *Salmonella* group B (O4, O5 , O27)
- *Salmonella* somatic (O) antiserum- *Salmonella* group C (O7, O8)
- *Salmonella* somatic (O) antiserum- *Salmonella* group D (O9, Vi)
- *Salmonella* somatic (O) antiserum - *Salmonella* group E (O3, O19)

- Anti- *Salmonella* flagella (H) e.g. e, f, g, h, i, k, l, m, p, q, r, s, t, u, v, w, x, z4, z23,z6, z29, z32, 1, 2, 5, 6, 7

APPENDIX B

MEDIA PRE PARAT ION

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

Modified Semi Solid Rappaport Visiliadis broth (MSRV; Merck KGaA, Germany)

Preparation: Suspend 42.5 g in 1 liter of autoclave water; **Do not** autoclave this medium, boiling, prior to use added MSRV supplement after that distributed into sterile Petri dishes 10 ml.

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.

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 heatingplate!).Cool to approx. 50 °C and pour plates. Do not overheat, do not autoclave!pH: 7.4 ± 0.2 at 25 °.

BPLS (BPLS; Merck KGaA, Germany)

Preparation: Suspend 51 g in 1 liter of demineralized water; boiling, autoclave (15 min. at 121 °C) pH: 7.0 ± 0.2 at 25 °C, plating.

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.

MIL (MIL; 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**

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**

APPENDIX C

COMPARISON OF *SALMONELLA* SEROTYPES IN THIS STUDY WITH THE OTHER STUDIES

Serogroup	Serotypes	Percentage		
		This study	Dorn-in (2005)	Sangwatanakul (2007)
B	*S.* Stanley	14.28	11.5	0
	S. Typhimurium	-	18.3	6.06
C	*S.* Rissen	57.14	45.4	18.18
E	-	-	14.6	-
D	-	-	2.0	-
II	F-67	25 (2/8)	3.7	75.76

DECLARATION

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

Name Ruttayaporn Ngasaman

Signature.....

Date of submission.....25 Sep 07.....

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