

BACTERIAL PATHOGENS ISOLATED FROM RAW MEAT AND POULTRY COMPARED WITH PATHOGENS ISOLATED FROM CHILDREN IN THE SAME AREA OF RURAL THAILAND

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Abstract. To better understand the epidemiology of bacterial food borne pathogens in children, in relation to pathogens in meats from a market in rural Thailand, we collected 73 cultures samples from raw chicken, pork and fish at a local market where diarrheal disease surveillance was conducted. Standard methods were employed to isolate, identify and serotype enteric pathogens from children and food samples. Antibiotic susceptibility testing was performed. Ninety-seven percent of food samples were contaminated with at least one enteric pathogen. The pathogens most commonly isolated from food were *Salmonella* spp (84%), *Arcobacter butzleri* (74%) and *Campylobacter* spp (51%). The most common serovars of *Salmonella* obtained from humans with diarrhea were *S. Risen*, *S. Stanley* and *S. Anatum*. Most common serovars of *Salmonella* isolated from food were *S. Anatum*, *S. Stanley*, and *S. Corvallis*. Fifty-one percent and 25% of children infected with *Salmonella* and *Campylobacter*, respectively, infected with the same serotypes isolated from food samples, suggesting these pathogens are widespread in food and humans. Pulsed-field gel analysis of *Salmonella* spp revealed 65 pulsotypes, but no point-sources of salmonellosis were identified. Joint epidemiologic/laboratory studies are useful to describe the epidemiology of enteric pathogens in rural populations.

Keywords: *Campylobacter*, *Salmonella*, *Arcobacter*, diarrhea, food, Thailand

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The views expressed in this publication are those of the authors and do not reflect the views of the US Department of the Army or Department of Defense.

INTRODUCTION

Consumption of contaminated food is an important risk factor for diarrheal illness, and is commonly identified as a source of outbreak transmission. The WHO Food Safety Unit estimated that in 2005, approximately 1.8 million children died of diarrheal disease; many of which were caused by contamination of food and drinking water (WHO, 2012).

Salmonella and *Campylobacter* species are major food borne pathogens causing diarrheal diseases worldwide (Butzler and Oosterom, 1991). *Salmonella enterica* is the most common *Salmonella* species causing human gastroenteritis while *C. jejuni* and *C. coli* are most frequently isolated from patients with *Campylobacter*-induced gastroenteritis (Butzler and Oosterom, 1991). Food of animal origin, especially meat and meat products, has been identified as a source of transmission of nontyphoidal *Salmonella* spp (Davies *et al*, 1998; Letellier *et al*, 1999; Limawongpranee *et al*, 1999; Bangtrakulnonth *et al*, 2004; Vindigni *et al*, 2007) and *Campylobacter* (Wilson, 2002; Vindigni *et al*, 2007). Human infections due to nontyphoidal *Salmonella* and *Campylobacter* generally occur through consumption of undercooked meat and meat products with fecal contamination or contaminated water or raw milk (WHO, 2007).

Aerotolerant *Campylobacter*-like organisms (CLO) have been reclassified as genus *Arcobacter* (Vandamme *et al*, 1991; Vandamme and Goossens, 1992; Winters and Slavik, 2002). Their ability to grow at lower temperatures and in air differentiates them from *Campylobacter* spp (Vandamme and Goossens, 1992). *Arcobacter* spp has been isolated from animals, sewage, poultry, and food in restaurants and some species are reported to be potentially pathogenic to humans (Morita *et al*, 2004; Vindigni *et al*, 2007; Teague *et al*, 2010).

In developing countries where enteric disease is endemic, point-source investigations of diarrheal illness are often hampered by the fact that foods implicated as vehicles of infection are not available for culture. An important aspect of research on disease transmission is identification of enteropathogens from food sources, and correlation of these isolates with those

from symptomatic patients. To date, such laboratory studies of enteric pathogens have been conducted only rarely in remote areas. A year-long case-control study of diarrheal disease to describe prevalences of enteric pathogens in children under five years of age was conducted at the Kwai River Christian Hospital (KRCH) in western Thailand, on the Thai-Myanmar border (Bodhidatta *et al*, 2010). The species, serotypes, serovars, antimicrobial susceptibilities and molecular typing of enteric pathogens isolated from children with and without diarrhea were compared to isolates from raw food samples purchased in the main local market.

MATERIALS AND METHODS

Sample collection and processing

Seventy-three culture samples obtained from raw chicken, pork, and fish were purchased from the local market in Nong Loo Subdistrict, Sangkhla Buri District, Kanchanaburi Province, Thailand every 3 months from October 2002 to July 2003. This market serves as a main food source for the populations of 6 villages located within a 9-km radius of where the diarrheal disease surveillance was conducted and described elsewhere (Bodhidatta *et al*, 2010). Raw food samples were processed according to the Bacteriological Analytical Manual (BAM) (Bacteriological Analytical Manual Online, 2001), US Food and Drug Administration/Center for Food Safety and Applied Nutrition (FDA/CFSAN), with some modifications as described below. Pieces of raw chicken, pork and fish, weighing at least 100 g, were collected in sterile zippered plastic bags. The bags were stored on ice from when they were obtained until they were delivered the same day to the Armed Forces Research Institute of Medical Sciences in Bangkok, Thailand.

Bacterial culture and identification

Salmonella was cultured with a combination of McConkey (MAC) and Modified semisolid Rappaport-Vassiliadis (MSRV) agar and Rappaport-Vassiliadis (RV) enrichment medium. The MAC and MSRV plates were incubated at 37°C for 18-24 hours and 42°C for 20-24 hours, respectively.

Campylobacter and *Arcobacter* were cultured with modified Charcoal-Cefoperazone Deoxycholate agar (mCCDA) and Bolton enrichment broth. Undiluted and diluted portions of Bolton broth were millipore filtered onto Brucella agar with 5% sheep blood (BAP). The plates were incubated for 48 hours at 37°C under microaerobic conditions (5% O₂, 10% CO₂, 85% N₂).

Shigella, *Aeromonas* and *Plesiomonas* were cultured with Hektoen Enteric (HE) and MAC agar. Enrichment was carried out on Shigella broth with novobiocin 0.5 µg/ml where it was incubated anaerobically at 42°C in a water bath for 20 hours followed by inoculation onto HE and MAC agar. The plates were incubated at 35°C for 20 hours.

Salmonella, *Aeromonas* and *Plesiomonas* species were subsequently identified and serotyped by standard microbiological methods. *Campylobacter* and *Arcobacter* were initially identified by colony characteristics, Gram stain and the oxidase test, followed by conventional phenotypic tests, including catalase, hippurate hydrolysis, nitrate reduction, indoxyl acetate hydrolysis, H₂S production in TSI, growth at 37°C aerobically, ability to grow at 25°C and 42°C microaerobically, and susceptibility to nalidixic acid (30 µg) and cephalothin (30 µg) disks (Lior *et al*, 1982; On *et al*, 1996; Isenberg, 1998; Murray *et al*, 1999).

Antibiotic susceptibility testing

Antibiotic susceptibility testing was performed by the disk diffusion method for all identified bacterial enteropathogens (Bauer *et al*, 1966; NCCLS, 2000, 2001). Susceptibility was tested against ampicillin (AM), trimethoprim/sulfamethoxazole (SXT), tetracycline (Te), nalidixic acid (NA), ciprofloxacin (CIP) and azithromycin (AZM). In the absence of NCCLS definitive standards for interpreting *Campylobacter* susceptibility results, established breakpoints for the family Enterobacteriaceae were used (NCCLS, 2001). For *Campylobacter* and other enteropathogens, azithromycin interpretive standards published by the disk manufacturer for *Staphylococcus* spp were used (BBL package insert).

Pulsed-field gel electrophoresis (PFGE)

PFGE was performed on 141 *Salmonella* isolates obtained from 15 children with diarrhea, 21 non-diarrhea controls and 61 food samples according to a standardized protocol previously described (CDC, 2004). The genomic DNA of the *Salmonella* isolates was extracted and digested with restriction enzyme *Xba*I. The samples of digested genomic DNA were run on CHEF Mapper systems (BioRad, Hercules, CA) and *Xba*I-digested DNA from *Salmonella* Braenderup was included as a standard marker. Gel was stained with ethidium bromide solution and visualized by a Gel documentation system (SynGene, Cambridge, UK). Band patterns were analyzed and dendrograms were generated using BioNumerics version 6.1 (Applied Maths, Saint Martin, Belgium).

Statistical analysis

Statistical analysis was performed to compare isolate resistance to antibiotics for *Salmonella* and *Campylobacter* from food and children by a two-tailed chi-

Table 1
Percent of enteropathogens among raw food samples in Sangkhla Buri, Thailand.

Enteropathogens	Total (n=73) No. (%)	Chicken (n=40) No. (%)	Pork (n=23) No. (%)	Fish (n=10) No. (%)
<i>Salmonella</i>	61 (84)	37 (93)	19 (83)	5 (50)
<i>Campylobacter</i>	37 (51)	32 (80)	4 (17)	1 (10)
<i>Plesiomonas</i>	20 (27)	12 (30)	3 (13)	5 (50)
<i>Arcobacter butzleri</i>	54 (74)	31 (78)	14 (61)	9 (90)
<i>Aeromonas</i>	4 (5)	1 (3)	1 (4)	2 (20)
No bacterial enteric pathogen detected	2 (3)	0 (0)	2 (9)	0 (0)

Table 2
Percent of multiple pathogens identified from food samples and stool samples from children with and without diarrhea in Sangkla Buri, Thailand.

Samples (n)	Enteropathogens identified		
	Single pathogen No. (%)	Multiple pathogens No. (%)	No pathogen No. (%)
Food (73)	7 (9.6)	64 (87.7)	2 (2.7)
Stool samples from children with diarrhea (236)	82 (34.7)	82 (34.7)	72 (30.6)
Stool samples from children without diarrhea (236)	100 (42.4)	60 (25.4)	76 (32.2)

square or Fisher's exact test. A *p*-value of <0.05 was considered statistically significant.

RESULTS

Seventy-three food samples were obtained: 40 samples were from chicken and internal organs, 23 samples were from pork and porcine internal organs and 10 samples were from fish. *Salmonella* (84%), *Campylobacter* (51%), and *Arcobacter butzleri* (74%) were the bacterial pathogens most commonly detected. *Campylobacter* was most commonly identified in chicken (32/40) and *Salmonella* was found in both chicken (37/40) and pork (19/23). *A. butzleri* was isolated from 31/40, 14/23 and

9/10 samples of chicken, pork and fish, respectively (Table 1). At least one bacterial enteric pathogen was detected in all except for 2 food samples (97%); the majority of them were contaminated with multiple pathogens or serotypes (Table 2).

Fifty-one percent (19/37) of preschool children were infected with *Salmonella* serotypes detected in food samples. The most common serovars from human isolates were *S. Risen*, *S. Stanley* and *S. Anatum*. The most common serovars from food isolates were *S. Anatum*, *S. Stanley* and *S. Corvallis* (Table 3). Children were infected with *C. jejuni* and *C. coli* of various Lior subtypes isolates (Table 4). Twenty-five percent (31/124) of human *C.*

COMPARISON OF ENTERIC PATHOGENS ISOLATED FROM RAW FOODS AND FROM CHILDREN

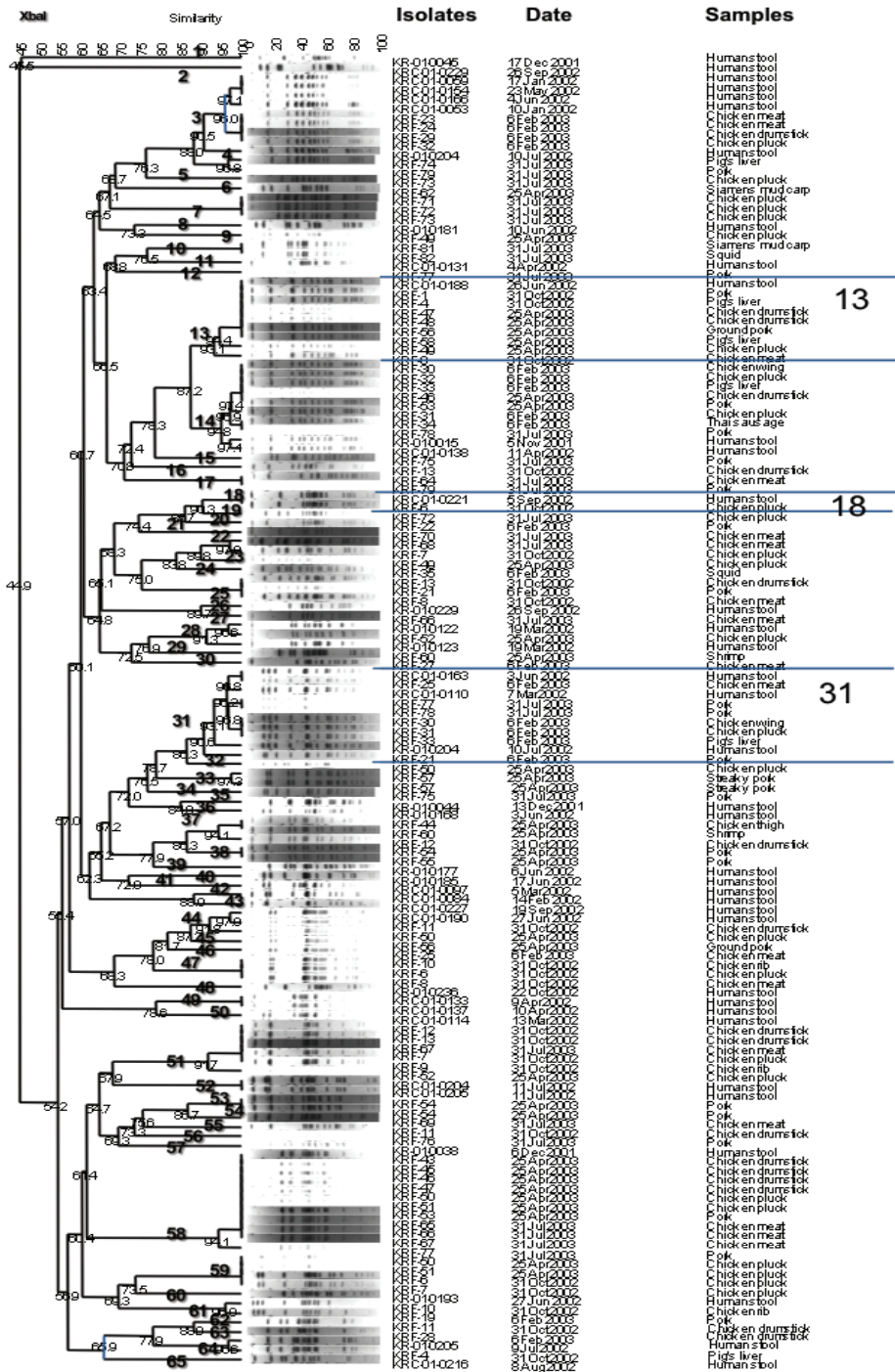


Fig 1—PFGE of *Salmonella* isolates from food and children in Sangkhla Buri, Thailand. Bold numbers in dendrogram are pulsotypes 1-65; three columns describe sample numbers, isolation date and sample types, respectively. Pulsotypes 13, 18 and 31 showed identical *Salmonella* serovars isolated from children and food samples.

Table 3

Major *Salmonella* serovars isolated from food and children in Sangkhla Buri, Thailand.

<i>Salmonella</i> serovars	Food (n=104) No. (%)	Children (n=37) No. (%)
S. Stanley	11 (11)	4 (11)
S. Anatum	16 (15)	3 (8)
S. Risen	6 (6)	5 (14)
S. Panama	8 (8)	1 (3)
S. Agona	3 (3)	1 (3)
S. Blockley	1 (1)	2 (5)
S. Hadar	7 (7)	1 (3)
S. Newport	2 (2)	1 (3)
S. Worthington	1 (1)	1 (3)
Subtotal	55 (53)	19 (51)

Other human isolates (18): *S. Bovis morbificans* (5), *S. Lexington* (2), *S. Weltevreden* (2), *S. Alachua* (1), *S. Orion* (1), *S. Altona* (1), *S. enterica* subsp *Enteritica* ser 4,5,12: (3), *S. enterica* subsp *Enteritica* ser 9,12: (1), *S. enterica* subsp *Houtenae* ser 43:24 (1), Serovar not tested (1). Other food isolates (49): *S. Corvallis* (11), *S. Enteritidis* (6), *S. Braenderup* (5), *S. Derby* (6), *S. Give* (4), *S. Virchow* (4), *S. Ruiru* (3), *S. Senftenberg* (3), *S. Albany* (2), *S. Paratyphi* (1), *S. Typhimurium* (1), *S. Montevideo* (1), *S. Idikan* (1), *S. Enteritica* (1).

jejuni and *C. coli* isolates belonged to the same Lior types as the isolates from food samples. *C. upsaliensis* and *C. doylei* were isolated in 19% (24/124) and 7% (9/124), respectively. These two *Campylobacter* spp were isolated from children but not from food samples.

PFGE of *Salmonella* isolates from children and food samples revealed 65 distinct pulsotypes (Fig 1). Identical clones (100% similarity) were observed among 72/142 *Salmonella* isolates belonging to 15 serovars (*S. Rissen*, *S. Ruiru*, *S. Newport*, *S. Anatum*, *S. Seftenberg*, *S. Hadar*,

Table 4

Campylobacter serotypes isolated from food and children in Sangkhla Buri, Thailand.

<i>Campylobacter</i> serotypes	Food (n=52) No. (%)	Children (n=124) No. (%)
<i>Campylobacter jejuni</i> Lior serotype		
114	5 (10)	3 (2)
36	3 (6)	14 (11)
5	2 (4)	0
11	2 (4)	0
28	1 (1)	2 (2)
4	0	12 (10)
7	0	5 (4)
1	0	3 (2)
9	0	3 (2)
102	0	3 (2)
2	0	1 (0.8)
19	0	1 (0.8)
94	0	2 (2)
doylei	0	9 (7)
Untypable <i>C. jejuni</i>	6 (11)	18 (15)
Subtotal for <i>C. jejuni</i>	19 (36)	76 (61)
<i>Campylobacter coli</i> Lior serotype		
55	8 (15)	2 (2)
46	6 (12)	2 (2)
21	5 (9)	0
8	4 (8)	5 (4)
29	2 (4)	1 (0.8)
72	2 (4)	2 (2)
110	0	4 (3)
44	0	1 (0.8)
Untypable <i>C. coli</i>	6 (12)	6 (5)
Subtotal for <i>C. coli</i>	33 (64)	23 (19)
<i>C. upsaliensis</i>	0	24 (19)
<i>Campylobacter</i> spp	0	1 (0.8)

S. Stanley, *S. Braenderup*, *S. Panama*, *S. Enteritidis*, *S. Lexington*, *S. Derby*, *S. Corvallis*, *S. Give* and *S. Altona*). Three pulsotypes (13, 18 and 31) (Table 7 and Fig 1) were the same serovar in both children and food samples with a 100% identical PFGE pattern.

Table 5
Antibiotic resistance among *Salmonella* isolates from food and children in Sangkhla Buri, Thailand.

	Food (n=104) No. (%)	Children (n=37) No. (%)	p-value
Ampicillin	22 (21)	10 (27)	NS
SXT ^a	25 (24)	8 (22)	NS
Tetracycline	60 (58)	18 (49)	NS
Nalidixic acid	45 (43)	7 (19)	0.0084
Ciprofloxacin	0 (0)	0 (0)	-
Azithromycin	1 (1)	2 (5)	-

^a SXT, trimethoprim/sulfamethoxazole

Table 6
Antibiotic resistance among *Campylobacter* isolates from food and children in Sangkhla Buri, Thailand.

	Food (n=52) No. (%)	Children (n=124) No. (%)	p-value
Ampicillin	14 (27)	14 (11)	0.009
SXT ^a	11 (21)	55 (44)	0.004
Tetracycline	33 (63)	27 (22)	< 0.00001
Nalidixic acid	51 (98)	57 (46)	< 0.00001
Ciprofloxacin	46 (88)	47 (38)	< 0.00001
Azithromycin	7 (13)	0	0.0002

^aSXT; trimethoprim/sulfamethoxazole

Salmonella isolates from children with and without diarrhea and food samples were all susceptible to ciprofloxacin and 76-78% of isolates were susceptible to SXT. Isolates from food were significantly more resistant to NA than isolates from children (Table 5). *Campylobacter* isolates from food were significantly more resistant than those from children to AM, Te, NA, CIP and AZM. In contrast, resistance to SXT was significantly higher among isolates from children than isolates from food samples (Table 6).

DISCUSSION

In this study, only 3% of raw food samples were found to be free of bacterial enteric pathogens while the five most common enteric bacteria contaminating food samples were *Salmonella*, *Arcobacter*, *Campylobacter*, *Plesiomonas* and *Aeromonas*. This proportion of contamination is much higher in comparison with previous studies from Thailand (Rasrinaul *et al*, 1988; Echeverria *et al*, 1994; Vindigni *et al*, 2007). This disparity could be explained

Table 7

Summary of pulsotypes, serogroups and serovars of *Salmonella* spp isolated from food and children in Sangkhla Buri, Thailand.

Pulsotypes	No. of isolates	Sources (n) ^a	Serogroup	Serovar	% Similarity
1	1	Human stool	SALD	NA ^c	
2	1	Human stool	SALD	ENTERICA SUBSP HOUTENAE SER 43:Z4,	
3	9	Human stool (4) Chicken meat (2) Chicken drumstick (1) Chicken pluck (1) Human stool (1)	SALC1 SALC SALC SALC SALC	RISEN (4) RISEN (2) RISEN (1) RISEN (1) RISEN (1)	90.5-100
4	2	Pig's liver Pork	SALC SALC	RISEN (1) DERBY	96.8
5	1	Chicken pluck	SALC	RISEN (1)	
6	1	Siamens mud carp	SALB	ENTERICA	
7	3	Chicken pluck Chicken pluck Chicken pluck	SALL SALL SALL	RUIRU RUIRU RUIRU	100
8	1	Human stool	SALB	ENTERICA SUBSP ENTERICA SER 4,5,12:	
9	1	Chicken pluck	SALB	TYPHIMURIUM	
10	2	Siamens mud carp Squid	SALC SALC	NEWPORT NEWPORT	100
11	1	Human stool	SALC	NEWPORT	
12	1	Pork	SALG	IDIKAN	
13	9	Human stool (1) ^b Pork (1) ^b Pig's liver (2) ^b Chicken drumstick ^b (2) Ground pork (1) ^b Chicken pluck (1) Chicken meat (1)	SALE SALE SALE SALE SALE SALE SALE	ANATUM ANATUM ANATUM ANATUM ANATUM ANATUM ANATUM	93.1-100
14	10	Chicken wing (1) Chicken pluck (2) Pig's liver (1) Chicken drumstick (1) Pork (2) Thai sausage (1) Human stool (1) Human stool (1)	SALE SALE SALE SALE SALE SALE SALE SALE1	ANATUM ANATUM ANATUM ANATUM ANATUM ANATUM ANATUM ANATUM	94.8-100
15	1	Pork	SALE	ANATUM	
16	1	Chicken drumstick	SALE	SENFENBERG	
17	2	Chicken meat Pork	SALE SALE	SENFENBERG SENFENBERG	100
18	2	Human stool ^b Chicken pluck ^b	SALC SALC	HADAR HADAR	100
19	1	Chicken pluck	SALC	HADAR	
20	1	Pork	SALC	HADAR	

Table 7 (Continued).

Pulsotypes	No. of isolates	Sources (n) ^a	Serogroup	Serovar	% Similarity
21	1	Chicken meat	SALC	HADAR	
22	2	Chicken meat	SALC	VIRCHHOW	97
		Chicken pluck	SALC	VIRCHHOW	
23	1	Chicken pluck	SALC	VIRCHHOW	
24	1	Squid	SALC	VIRCHHOW	
25	3	Chicken drumstick	SALC	HADAR	100
		Pork	SALC	HADAR	
		Chicken meat	SALC	HADAR	
26	1	Human stool	SALG	WORTHINGTON	
27	1	Chickens meat	SALG	WORTHINGTON	
28	3	Human stool	SALC	BLOCKLEY	91.3-96.6
		Chicken pluck	SALC	BLOCKLEY	
		Human stool	SALC	BLOCKLEY	
29	1	Shrimp	SALD	PANAMA	
30	1	Chicken meat	SALB	PARATYPHI	
31	10	Human stool ^b	SALB	STANLEY	90.6-100
		Chicken meat ^b	SALB	STANLEY	
		Human stool	SALB	STANLEY	
		Pork	SALB	STANLEY	
		Pork	SALB	STANLEY	
		Chicken wing	SALB	STANLEY	
		Chicken pluck	SALB	STANLEY	
		Pig's liver	SALB	STANLEY	
		Human stool	SALB	STANLEY	
		Pork	SALB	STANLEY	
32	1	Chicken pluck	SALB	STANLEY	
33	2	Streaky pork	SALB	STANLEY	97.3
		Streaky pork	SALB	STANLEY	
34	1	Pork	SALB	STANLEY	
35	1	Human stool	SALB	ENTERICA SUBSP ENTERICA SER 4, 5, 12:	
36	1	Human stool	SALB	ENTERICA SUBSP ENTERICA SER 4, 5, 12:	
37	3	Chicken thigh	SALC	BRAENDERUP	94.1-100
		Shrimp	SALC	BRAENDERUP	
		Chicken drum	SALC	BRAENDERUP	
38	2	Pork	SALC	BRAENDERUP	100
		Pork	SALC	BRAENDERUP	
39	1	Human stool	SALB	STANLEY	
40	1	Human stool	SALC	BOVISMORBIFICANCE	
41	1	Human stool	SALC	BOVISMORBIFICANCE	
42	1	Human stool	SALE	WELTEVREDEN	
43	1	Human stool	SALE	WELTEVREDEN	
44	3	Human stool	SALD	PANAMA	91.8-97
		Chicken drumstick	SALD	PANAMA	
		Chicken pluck	SALD	PANAMA	
45	1	Ground pork	SALD	PANAMA	

Table 7 (Continued).

Pulsotypes	No. of isolates	Sources (<i>n</i>) ^a	Serogroup	Serovar	% Similarity
46	1	Chicken meat	SALD	PANAMA	
47	3	Chicken rib	SALD	PANAMA	100
		Chicken pluck	SALD	PANAMA	
		Chicken meat	SALD	PANAMA	
48	1	Human stool	SALB	ENTERICA SUBSP ENTERICA SER 9,12:-:	
49	2	Human stool	SALC	BOVISMORBIFICANCE	100
		Human stool	SALC	BOVISMORBIFICANCE	
50	1	Human stool	SALC	BOVISMORBIFICANCE	
51	6	Chicken drumstick	SALD	ENTERITIDIS	91.7-100
		Chicken drumstick	SALD	ENTERITIDIS	
		Chicken meat	SALD	ENTERITIDIS	
		Chicken pluck (2)	SALD	ENTERITIDIS	
		Chicken rib	SALD	ENTARITIDIS	
52	2	Human stool	SALE	LEXINGTON	100
		Human stool	SALE	LEXINGTON	
53	2	Pork	SALB	DERBY	100
		Pork	SALB	DERBY	
54	1	Chicken meat	SALB	DERBY	
55	1	Chicken drumstick	SALC	MONTEVIDE	
56	1	Pork	SALB	DERBY	
57	1	Human stool	SALE	ORION VAR 15+	
58	11	Chicken drumstick	SALC	CORVALIS	94.1-100
		Chicken drumstick	SALC	CORVALIS	
		Chicken drumstick	SALC	CORVALIS	
		Chicken drumstick	SALC	CORVALIS	
		Chicken pluck	SALC	CORVALIS	
		Chicken pluck	SALC	CORVALIS	
		Pork	SALC	CORVALIS	
		Chicken meat	SALC	CORVALIS	
		Chicken meat	SALC	CORVALIS	
		Chicken meat	SALC	CORVALIS	
		Pork	SALC	CORVALIS	
59	4	Chicken pluck	SALE	GIVE	100
		Chicken pluck	SALE	GIVE	
		Chicken pluck	SALE	GIVE	
		Chicken pluck	SALE	GIVE	
60	1	Human stool	SALO	ALACHUA	
61	2	Chicken rib	SALC	ALBANY	96
		Pork	SALC	ALBANY	
62	1	Chicken drumstick	SALB	AGONA	
63	1	Chicken drumstick	SALB	AGONA	
64	2	Human stool	SALB	AGONA	96.6
		Pig's liver	SALB	AGONA	
65	12	Human stool	SALC	ALTONA	

^a(*n*), number of isolates; ^bisolates from human (children) stool and food of 100% identity and same serovar; ^cNA, not applicable.

partially by the fact that *Arcobacter butzleri*, an “underestimated enteropathogen” as described by Prouzet-Mauléon *et al* (2006) had not been assessed for in other food surveys with the exception of Vindigni *et al* (2007). We detected *A. butzleri* in 1% each of children with and without diarrhea and in 74% of food samples. This disparity between human and food sources for *Arcobacter* needs further study.

Salmonella and *Campylobacter* were the two common pathogens isolated from food samples in this study. There was a 51% and 25% overlap in *Salmonella* and *Campylobacter* serotypes obtained from children and food in our study suggesting a common exposure for these two pathogens among pre-school children by food origin. These bacterial species were the two most common pathogens isolated in a similarly study from Bangkok in 1986 (Rasrinaul *et al*, 1988). However, one major difference between Rasrinaul *et al* (1988) and our study is the source of food items cultured. In our study, raw meat samples were collected from one local food market in a remote area where the children’s food was most likely purchased. Rasrinaul *et al* (1988) sampled meat and vegetable products from 10 different markets in Bangkok.

In a review of serovar trends in *Salmonella* isolates collected in human between 1993 and 2002 by the World Health Organization National Salmonella and Shigella Center in Bangkok, Thailand, and *Salmonella* isolates collected from food and water during the same period, *S. Weltevreden* (12.5/59.4%), *S. Enteritidis* (11.4/28%) and *S. Anatum* (7.4/33.4%) were the most common isolates (Bangtrakulnonth *et al*, 2004). Boonmar *et al* (1998a) found serovars differed by food item. The frequencies of isolation of *S. Enteritidis*, *S. Weltevreden*, *S. Derby* and *S. Anatum* in humans and in food items differed (Boon-

mar *et al*, 1998a). In another study from Thailand during 1990-1997, a close correlation was seen between *S. Enteritidis* isolates collected from humans and broiler meat products based on phage typing and pulsed-field gel electrophoresis (Boonmar *et al*, 1998b). Our study reveals many of the same *Salmonella* serovars isolated from children were also found in food items at the local market. The serovar distribution was different in another study from Thailand conducted during 2002-2007 that showed an increase in *S. Choleraesuis* but a decrease in *S. Weltevreden* in human isolates (Hendriksen *et al*, 2009). Our results find neither of these two serovars among children or food samples in Sangkhla Buri.

PFGE demonstrated heterogeneity of *Salmonella* spp isolated from food and children in Sangkhla Buri. However, the finding of identical clones possessed by 15 different serovars suggests these isolates were genetically related. Identical clones were observed in human and food isolates for three serovars: *S. Anatum* and *S. Hardar* and *S. Stanley*, which suggests a common distribution of these *Salmonella* serovars in humans and in food, but not as a source of infection.

A study comparing *Campylobacter* strains isolated from raw meat to strains isolated from humans reported 20 distinct subtypes of *C. jejuni* were isolated from both poultry and human fecal samples; the most common subtype isolated in humans was also the most common subtype isolated from poultry (Kramer *et al*, 2000). In our study, when comparing *Campylobacter* serotypes isolated from subjects and food, 25% shared the same Lior subtypes, However, *C. upsaliensis* and *C. jejuni* subspecies *doylei*, which accounted for 26% of all *Campylobacter* isolated from subjects was not found in food samples.

Antimicrobial resistance among food borne bacterial pathogens tends to occur in food/animals before it occurs in humans (White *et al*, 2002). In our study, *Campylobacter* isolates from food were significantly more resistant to antimicrobials than those from subjects for 5 out of 6 antibiotics tested. This did not hold true for SXT. This may be attributable in part to the wide availability and affordability of this drug in the study area. This drug is not commonly used in poultry from which most of the *Campylobacter* isolates were identified. Among *Salmonella* isolates, the pattern of greater resistance among isolates from food compared to those from children was seen in only 1 of 6 antibiotics tested: nalidixic acid.

The presumed source of resistance is the use of antibiotics in animal-production (White *et al*, 2001). Several authors have recommend guidelines be established to restrict the use of fluoroquinolones in animals, limiting dissemination of fluoroquinolone-resistant *Salmonella* (Angulo *et al*, 2000; Zhao *et al*, 2003) and *Campylobacter* (Witte, 1998; Pezzotti *et al*, 2003) to human. Interventions, such as selective vaccination of broiler flocks (Wilson, 2002) and bans on antimicrobial growth promoters at sub-therapeutic doses for animals (Wierup, 2001) have resulted in decreases in the incidence of contamination of food sources in Ireland and Sweden, respectively.

According to the Centers for Disease Control Food Net Surveillance Network (CDC, 2006), the most important source of *Salmonella* infection is food animals. Transmission can occur by ingestion of contaminated food sources or by direct contact with contaminated animals. Measures aimed at preventing human infection can be specifically modeled when the source of infection is known. Hence, reducing the

risk of food borne illness requires identifying animal reservoirs and transmission routes (CDC, 2006). This can be expanded to other food borne illnesses identified in our study population. We have attempted to identify the food sources of bacterial enteropathogens in Sangkhla Buri, Thailand, and drawn comparisons between these isolates and those in humans. Such epidemiologic data is important for preventive strategies and control of diarrheal diseases, especially in remote areas where populations share food sources available in only a few local markets.

ACKNOWLEDGEMENTS

We are thankful to the staff of the Enteric Diseases Department, AFRIMS, Bangkok, for bacterial identification, collection and molecular typing. This work was supported by the Armed Forces Health Surveillance Center-Global Emerging Infections Surveillance and Response System (AFHSC-GEIS), Washington, DC, USA.

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