C-EnterNet 2009 Annual Report

...National Integrated Enteric Pathogen Surveillance Program

Canada

To promote and protect the health of Canadians through leadership, partnership, innovation and action in public health.

— Public Health Agency of Canada

C-EnterNet 2009 Annual Report

...National Integrated Enteric Pathogen Surveillance Program

Acknowledgements

C-EnterNet Advisory Committee (cont'd)

Deborah Yamamura, LifeLabs
Chris Green, Manitoba Agriculture, Food and Rural Interests
Eduardo Taboada, Laboratory for Foodborne Zoonoses, Public Health Agency of Canada
Dylan Pillai, Ontario Agency for Health Protection and Promotion,
 Public Health Laboratories Toronto

B.C. Centre for Disease Control

Eleni Galanis, Marsha Taylor

BCCDC Public Health Microbiology and Reference Laboratory

Judith Isaac-Renton

Canadian Food Inspection Agency
Canadian Medical Laboratories

Maureen Lo, Phil Stuart, Maria Suglio

Canadian Public Health Laboratory Network
Gamma-Dynacare Laboratories

Kathy Biers, Julius Kapala

Grand River Conservation Authority

Mark Anderson

Grand River Hospital - Regional Microbiology Laboratory

John Vanderlaan

Health Canada - Bureau of Microbial Hazards

Sabah Bidawid, Brent Dixon, Jeff Farber, Karine Hebert, Kirsten Mattison, Oksana
Mykytczuk, Franco Pagotto, Lorna Parrington, Anu Shukla, Kevin Tyler

Hyperion Research Ltd.

Quynh Nguyen, Peter Wallis

LifeLabs

Colette Béchard

Ontario Agency for Health Protection and Promotion

Surveillance and Epidemiology Section
Dean Middleton

Public Health Laboratories - Toronto
Vanessa Allen, Peter Boleszchuk, Donald Low, Anne Maki, Dylan Pillai

Ontario Ministry of Agriculture, Food and Rural Affairs
Ontario Ministry of Environment

Deb Conrod, Wolfgang Scheider, Janis Thomas, David Supper

Suggested citation

Government of Canada. Canadian National Enteric Pathogen Surveillance System (C-EnterNet) 2011. Guelph, ON: Public Health Agency of Canada.

Executive Summary

C-EnterNet is a multi-partner sentinel site surveillance program facilitated by the Public Health Agency of Canada. Its core objectives are to: 1) detect changes in trends in human enteric disease and in levels of pathogen exposure from food, animal and water sources in a defined population; 2) generate human illness attribution values (proportion of human cases due to exposure via water, food and animals); and 3) improve the analysis, interpretation and reporting of laboratory and epidemiological data for public health, water and agri-food purposes.

Each sentinel site is based on a unique partnership with the local public health unit, private laboratories, water and agri-food sectors, as well as the provincial and federal institutions responsible for public health, food safety and water safety. The first sentinel site – the Region of Waterloo, Ontario – has approximately 500,000 residents, with a mix of urban and rural communities, and innovation in public health and water conservation. A second site was officially launched in the Fraser Health Authority, British Columbia in June 2010.

The following key messages have been developed based on the surveillance data from 2009 in Sentinel Site 1.

- Travel outside Canada continued to add to the burden of diseases observed in Canada with 29% of the reported cases likely infected abroad in 2009. Travel medicine practitioners should continue to promote safe travel practices to Canadians.

- While fewer endemic cases of enteric disease were reported in 2009 compared to 2008, incidence rates have been relatively stable over the last 4 years for most of these diseases. Exceptions were the rates of *E.coli* O157:H7 infections and yersiniosis that have significantly decreased since 2006.

- Campylobacteriosis remained the most common reported endemic disease and *Campylobacter jejuni* was the most common species associated with human infections. *Campylobacter jejuni* was also the most commonly detected type of *Campylobacter* detected on raw chicken breasts purchased at retail. Raw chicken demonstrated the greatest potential as vehicle for *Campylobacter* infections of the pathways examined highlighting the importance of safe cooking and food handling practices. Other pathways are also important as *Campylobacter* was also detected in environmental samples and contact with household pets was higher for the campylobacteriosis cases than the other cases.

- Salmonellosis was the second most frequently reported endemic disease. In food at retail, *Salmonella* was commonly detected in raw chicken, rarely in beef and pork and not in bagged leafy green. The frequency of contamination of raw chicken has been stable over time since 2006. *Salmonella* was also detected in samples from broiler, swine, beef, and dairy farms and in untreated surface water, with a slight increasing trend over the years. Chicken appeared to be a primary reservoir for *Salmonella* causing human illness as demonstrated by similarity of subtypes (e.g. *Salmonella* Enteritidis phagetypes 8, 13 and 13a) predominating in human cases, retail chicken meat and on chicken farms. Exposure to pet reptiles was higher for salmonellosis than the other diseases again in 2009

- The incidence of *E.coli* O157:H7 infections continued to decrease in 2009. The *E.coli* O157:H7 infections occurred during summer time. Multiple risk factors were reported more frequently in these cases compared to the other diseases including travel within Canada and outdoor activities such as canoeing, kayaking, hiking or camping. Pathogenic *E.coli* O157:H7 was found on beef and dairy farms. VTEC was found in one retail ground beef sample but not in bagged leafy greens or untreated surface water. These results indicated that cattle are a major reservoir for *E. coli* O157:H7 but there are multiple pathways for infection.

- The incidence rate of yersiniosis has continued a downward trend in 2009. They were primarily domestically-acquired infections with no seasonality. Pathogenic *Yersinia entercolitica* was found rarely on retail pork and swine farms but not in untreated surface water.

- One case of listeriosis was reported in the sentinel site in 2009. Pathogenic subtypes of *Listeria monocytogenes* were found on raw pork, chicken and beef as well as bagged leafy greens.

- Both *Giardia* and *Cryptosporidium* (including human-infectious and non-human-infectious strains) were routinely found in the surface water. They were also found in bagged leafy greens.

- Norovirus and rotavirus were found in bagged leafy greens by molecular techniques.

- Enhanced, standardized laboratory testing across all C-EnterNet components (human, farm, retail and water) has allowed for the identification of patterns in subtype distributions among human cases and potential exposure sources over time. Continued surveillance and the addition of more sentinel sites will help refine the key messages and inform prevention and control measures for enteric diseases in Canada.

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1. Introduction

C-EnterNet is a multi-partner sentinel site surveillance program facilitated by the Public Health Agency of Canada. Its core objectives are to: 1) detect changes in trends in human enteric disease and in levels of pathogen exposure from food, animal and water sources in a defined population; 2) generate human illness attribution values (proportion of human cases due to exposure via water, food and animals); and 3) improve the analysis, interpretation and reporting of laboratory and epidemiological data for public health, water and agri-food purposes.

Each sentinel site is based on a unique partnership with the local public health unit, private laboratories, water and agri-food sectors, as well as the provincial and federal institutions responsible for public health, food safety and water safety. The first sentinel site – the Region of Waterloo, Ontario – has approximately 500,000 residents, with a mix of urban and rural communities, and innovation in public health and water conservation. A second site was officially launched in the Fraser Health Authority, British Columbia in June 2010.

C-EnterNet conducts continuous and episodic surveillance activities in four components: human, food, water, and food animals. For a description of the suite of pathogen testing see Appendix A. Continuous surveillance occurs throughout the year to identify trends in human disease occurrence, exposure sources and source attribution for eleven enteric pathogens. Episodic surveillance activities are limited in time and provide specific information to complement the continuous activities. Detailed descriptions of the C-EnterNet design, laboratory methods and the enteric disease case questionnaires, are available at our website (http://www.phac-aspc.gc.ca/c-enternet/index-eng.php).

As in previous years, the 2009 report begins with a summary of the reported infectious enteric disease cases in humans in Sentinel Site 1, summarizing the outbreak- and travel-related cases separately from the endemic cases (Chapter 2). Chapters 3 through 10 provide information on human cases and exposure source surveillance for 2009 by pathogen, as in previous years. New to 2009, a *Shigella* and a virus chapter have been added, due to expanded exposure source monitoring for these pathogens in the retail food component. Chapter 11 provides a discussion of the temporal variations observed in the human cases and among the potential exposure sources from mid-2005 to the end of 2009. All observations and analyses dealing with trends and seasonality are addressed in this section. A summary of C-EnterNet's ongoing efforts to test and refine methodologies to estimate human illness attribution are presented in Chapter 12.

The surveillance data provided in this report only relate to the first sentinel site. Therefore, the accuracy of generalizing these results beyond this community decreases when moving further from the specific geographical area. As additional sentinel sites are implemented, comprehensive information from laboratory and epidemiological data from all sites will provide more representative national trends in enteric disease occurrence and among exposure sources. This will ultimately provide more accurate human illness attribution estimates for Canada.

2. Human Case Summary

2.1 Overview of Human Cases

A total of 391 cases of 11 bacterial, viral and parasitic enteric diseases were reported to the local public authorities within Sentinel Site 1 in 2009 (Table 2.1). The three most frequently reported diseases (salmonellosis, campylobacteriosis and giardiasis) accounted for 81% of those cases (Figure 2.1).

Risk Factor*: possible exposure source in the transmission of infection, such as consumption of a contaminated food or exposure to an animal*

Information on potential risk factors was obtained from 86% (of the 7 pathogens included in this report) of the reported cases within the sentinel site in 2009. Public health inspectors administered a standardized questionnaire to the cases or proxy respondents. Preliminary analyses of this information were used to determine case status (international travel versus endemic (domestic)) and compare risk factors (Appendix A).

- Outbreak-related: defined by public health partner (Sentinel Site 1) based on laboratory or epidemiological evidence

- International travel-related: case has traveled outside Canada prior to onset of illness, and the expected incubation period overlapped with the travel time

- Endemic: case is considered sporadic and domestically-acquired (in Canada)

TABLE 2.1
Number of cases and incidence rates per 100,000 person-years of laboratory-confirmed enteric diseases in Sentinel Site 1 in 2009

Disease	Incubation Period	Number of Cases				Incidence Rate	
		Outbreak	Travel	Endemic	Total	Endemic	Total
Amoebiasis	2-4 weeks	0	14	13	**27**	2.51	**5.21**
Campylobacteriosis	1-10 days	0	19	99	**118**	19.10	**22.76**
Cryptosporidiosis	1-12 days	0	3	17	**20**	3.28	**3.86**
Cyclosporiasis	2-14 days	0	4	0	**4**	0.00	**0.77**
Giardiasis	3-25 days	0	32	40	**72**	7.72	**13.88**
Hepatitis A	15-50 days	0	4	2	**6**	0.39	**1.16**
Listeriosis	3-70 days	0	0	1	**1**	0.19	**0.19**
Salmonellosis	6-73 hours	0	35	82	**117**	15.82	**22.57**
Shigellosis	1-3 days	0	1	7	**8**	1.35	**1.54**
Verotoxigenic *E. coli* (VTEC)	2-10 days	0	0	10	**10**	1.93	**1.93**
Yersiniosis	3-7 days	0	1	7	**8**	1.35	**1.54**
Total		**0**	**113**	**278**	**391**		
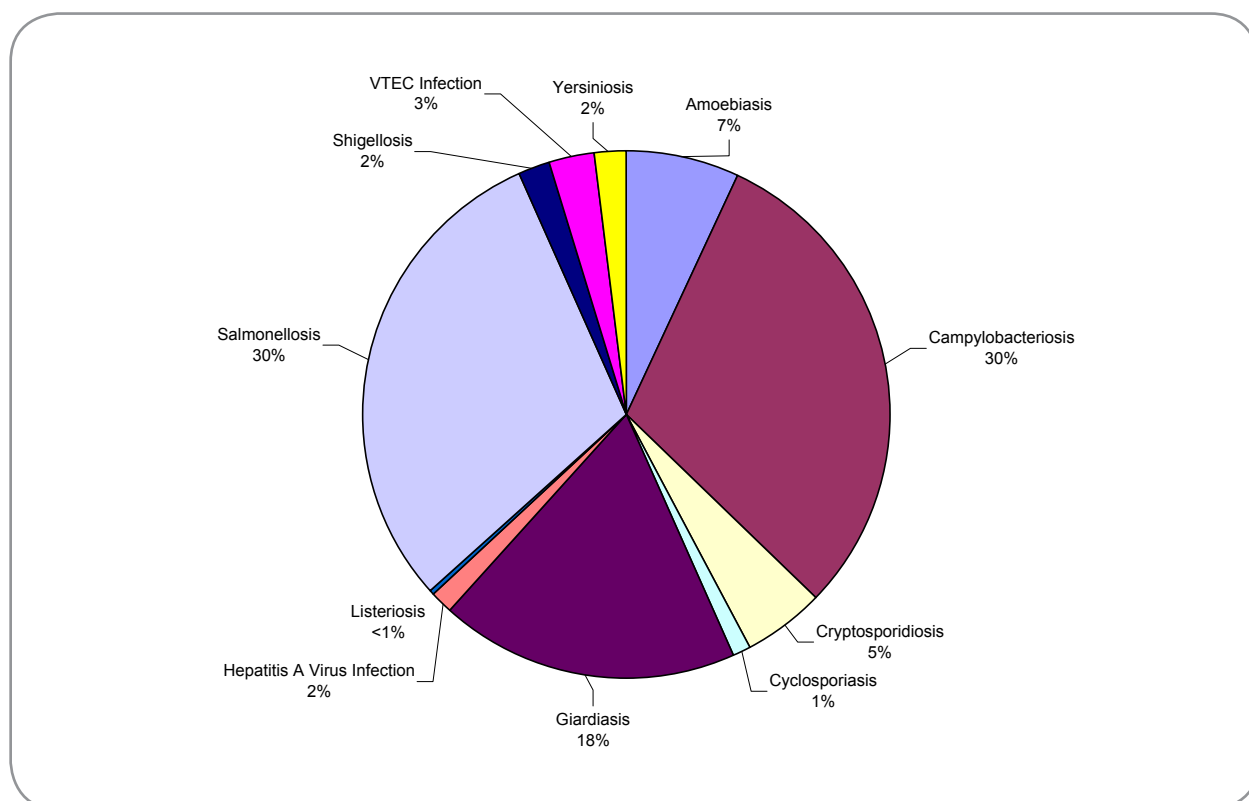
Figure 2.1. Relative proportion of enteric diseases reported in Sentinel Site 1 in 2009 (all cases)

From a clinical perspective, it is useful to record the site of isolation for each pathogen. The most common sample type is stool. Among *Salmonella* cases, the 6 human cases where the pathogen was detected from blood included serotypes Bonariensis, Heidelberg, Paratyphi A, ssp. I:4,5,12:B:-, and Typhi. *Shigella boydii* was isolated from blood from individual.

TABLE 2.2
Site of isolation of pathogens detected in laboratory-confirmed cases reported in Sentinel Site 1 in 2009 (including outbreak, international travel-related and endemic).

Disease	Site of Isolation				
	Blood	Stool	Urine	Unknown	Total
Amoebiasis	1	26	0	0	27
Campylobacteriosis	0	117	0	1	118
Cryptosporidiosis	0	20	0	0	20
Cyclosporiasis	0	3	0	1	4
Giardiasis	0	72	0	0	72
Hepatitis A	6	0	0	0	6
Listeriosis	0	0	1	0	1
Salmonellosis	6	106	4	1	117
Shigellosis	1	7	0	0	8
Verotoxigenic *E. coli* (VTEC)	0	10	0	0	10
Yersiniosis	0	8	0	0	8
Total	**14**	**369**	**5**	**3**	391

2.2   Outbreak-related Cases

In 2009, there were no outbreak-associated enteric disease cases (in the community, not associated with an institution).

In 2009, 61 institutional enteric outbreaks were identified and investigated. Twenty-seven outbreaks occurred in childcare centres (CCC), 26 in long-term care facilities (LTCF), 5 in residential facilities/group homes and 3 in hospitals. A causative agent was identified in 27% of outbreaks in LTCF and residential facilities/group homes combined and 7% of outbreaks in CCC. In LTCF and residential facilities/group home outbreaks, where the causative agent was identified, outbreaks were linked to norovirus and rotavirus, whereas rotavirus was identified in CCC.

2.3   Travel-related Cases

Travel-related cases: *individuals that have travelled outside of Canada in the relevant time frame before onset of illness.*

Of the reported cases, 29% (113/391) were classified as international travel-related (Table 2.1). Salmonellosis, giardiasis and campylobacteriosis were the three most common diseases, contributing to 76% of the travel-related cases. Most of the cases had visited Mexico and the Caribbean region or Asia prior to acquiring their illness (Table 2.3); a trend that possibly reflects travel preferences of the sentinel site population. Over half of the travel-related *Salmonella* cases, (21/35), had been to Mexico and the Caribbean region whereas giardiasis was the most frequent disease in patients who had travelled to Asia (17/36) and Africa (8/21). There were no travel-associated VTEC infections reported in 2009. *E.coli* O157:H7 continues to present as a domestically acquired infection in this community.

TABLE 2.3
Travel-related cases in Sentinel Site 1 in 2009

Disease	Africa	Mexico & Caribbean	Asia	Europe	USA	Multiple Destinations & Others	Total
Amoebiasis	5	4	5	0	0	0	**14 (12%)**
Campylobacteriosis	3	4	3	3	3	3	**19 (17%)**
Cryptosporidiosis	0	2	1	0	0	0	**3 (3%)**
Cyclosporiasis	0	2	1	0	1	0	**4 (3.5%)**
Giardiasis	8	6	17	0	1	0	**32 (28%)**
Hepatitis A	3	1	0	0	0	0	**4 (3.5%)**
Salmonellosis	2	21	8	3	1	0	**35 (31%)**
Shigellosis	0	0	1	0	0	0	**1 (1%)**
Verotoxigenic *E. coli*	0	0	0	0	0	0	**0 (0%)**
Yersiniosis	0	1	0	0	0	0	**1 (1%)**
Total	21 (19%)	41 (36%)	36 (32%)	6 (5%)	6 (5%)	3 (3%)	**113 (100%)**

2.4   Endemic Cases

Endemic cases*: reported cases of infection that occur sporadically within the sentinel site area (domestically acquired).*

The analyses presented in the remainder of this report largely refer to endemic cases. While outbreak cases are also attributed to local sources of exposure, they represent unusual events. By excluding outbreak and international travel cases, more stable estimates of disease incidence are provided, and attribution estimates will not be overly influenced by unusual events. Note that reported national and provincial annual incidence rates for each pathogen include both endemic, outbreak and travel cases and are from 2008, since the 2009 rates were not available at the time of publication. Although C-EnterNet is not actively monitoring pathogen exposure in other potential sources (such as pet animals), these risk factors are explored through the human case follow-up questionnaire used by the local health unit.

In each of the following sections, potential risk factors are noted when the proportion for the specific disease is at least 5% greater than the risk factor for other enteric diseases combined. Due to the small number of cases in Sentinel Site 1, exposure information was not stratified by age or gender. Thus, the risk factors reported here represent overall exposures for the general population, and are not valid for age-specific subgroups (e.g. children). Refer to the C-EnterNet website (http://www.phac-aspc.gc.ca/c-enternet/index-eng.php) to see the complete list of risk factors from the worksheet (questionnaire) used in Sentinel Site 1 for case follow-up investigations.

3. *Campylobacter*

3.1 Human Cases

In 2009, there were a total of 118 (22.8/100,000 person-years) reported cases of *Campylobacter* infection. Of these 118 cases, 16% (19/118) were travel-related (3.7/100,000 person-years), and 84% (99/118) were classified as endemic (19.1/100,000 person-years). In comparison, the annual incidence rates for campylobacteriosis in 2008 in Canada and Ontario were 28.4/100,000 and 29.4/100,000, respectively (**1**).

The age- and gender-specific endemic incidence rates were highest in males less than 5 years of age (46.3/100,000) and between 20-24 years of age (39.2/100,000) (Figure 3.1). A breakdown by gender shows that 53 cases were female (20.4/100,000) and 65 were male (25.1/100,000).
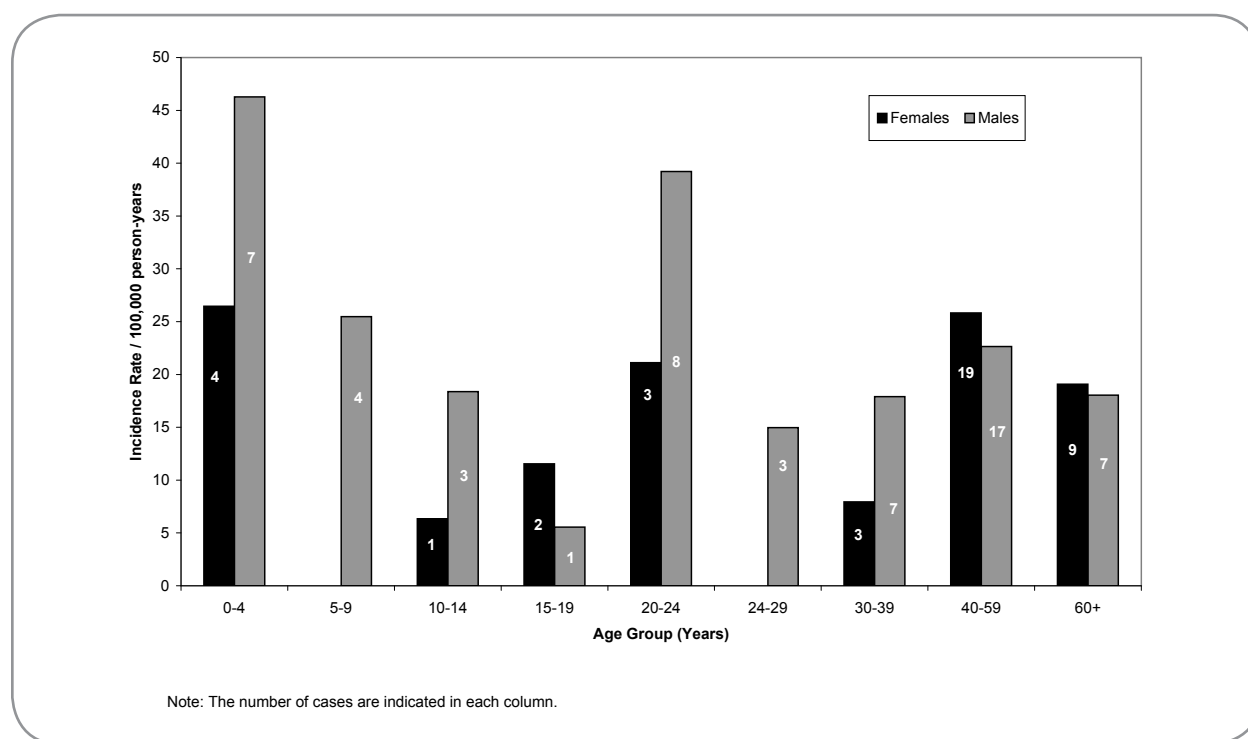
Figure 3.1. Incidence rates of endemic campylobacteriosis in Sentinel Site 1 by gender and age group in 2009

The majority (98%) of endemic campylobacteriosis cases were identified as *C. jejuni* while *C. coli* accounted for the remaining 2% (Table 3.1).
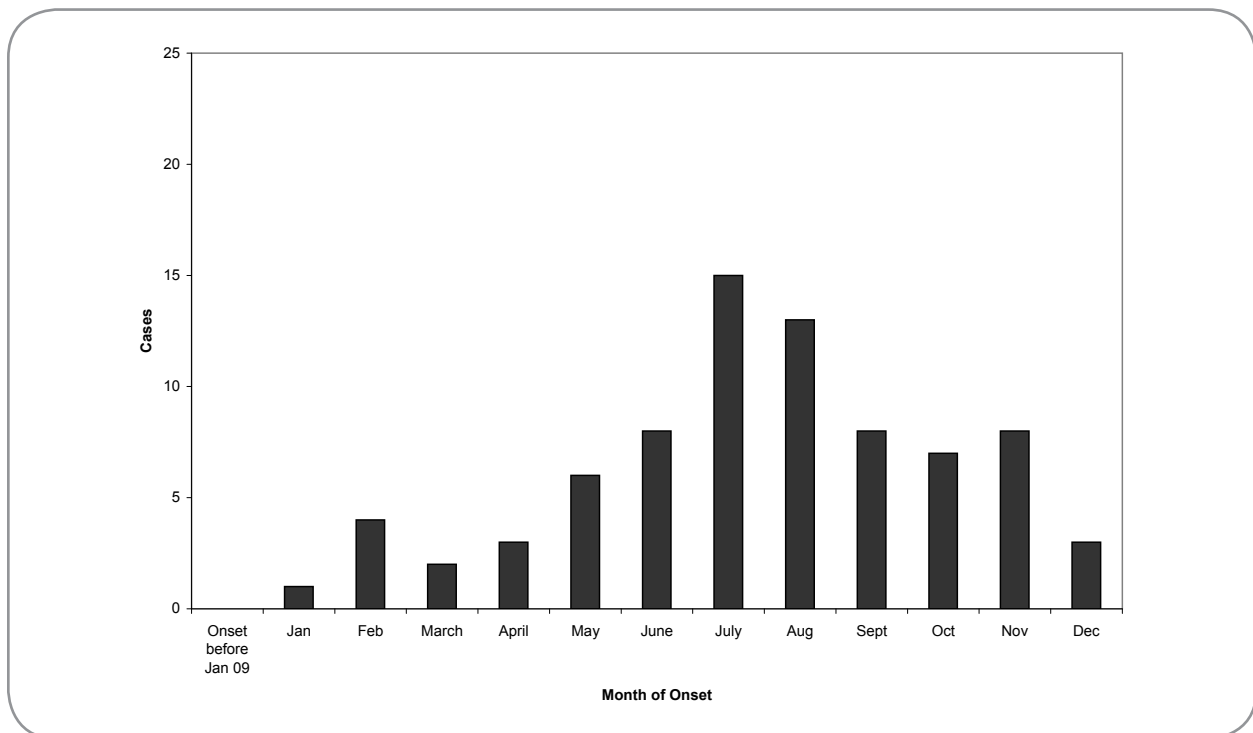
Figure 3.2. Temporal distribution of human endemic *Campylobacter* cases in Sentinel Site 1 reported in 2009 by month of onset

Eighty-one percent (80/99) of the endemic *Campylobacter* cases provided potential risk factor information for the 10 days prior to onset of illness (Appendix B). Use of municipal water source (66%), eating undercooked food (11%), attending a barbeque (26%), eating in a restaurant (28%), and visiting a farm animal area (16%) were reported more frequently among *Campylobacter* cases than among other enteric cases. *Campylobacter* cases had a higher proportion of household pet contact (58%), especially with cats (23%) and dogs (46%) than other enteric cases.

The incidence rate of endemic campylobacteriosis was higher during the summer months (June, July, August) than during the spring (March, April, May) [Fisher's Exact Test: $p < 0.05$] and winter (December, January, February) [Chi squared test: $p < 0.05$].

TABLE 3.1
**Campylobacter detection and speciation data (culture-based methods)
from integrated surveillance activities in Sentinel Site 1 in 2009**

Detection	Human	Retail Food				Food Animals (Manure)				Untreated Surface Water
	Endemic Cases	Pork	Chicken	Beef	Produce	Swine	Broiler Chickens	Beef Cattle	Dairy Cattle	Grand River
		Pork chop	Chicken breast	Ground beef	Bagged leafy greens	30 Farms	30 Farms	30 Farms	30 Farms	5 sample points on Grand River
# tested	Unknown	200	200	200	376	120	120	120	120	112
# positive	99	1	92	1	0	96 (30 farms)	6 (2 farms)	95 (30 farms)	96 (30 farms)	24
% positive		0.5%	46%	0.5%	0.0%	80%	5%	79%	80%	21%
Subtyping										
# subtyped	99	1	92	1		96	6	95	96	24
C. coli	2 2%	1 100%	6 7%			78 81%		14 15%	9 9%	5 (A,B,D) 21%
C. jejuni	97 98%		86 93%	1 100%		3 3%	6 100%	66 69%	69 72%	12 (A,B,C,D) 50%
C. lari										7 (A,B,C) 29%
C. upsaliensis										
Other						15 16%		15 16%	18 19%	

Water Sampling Locations in Grand River Watershed:
A - Canagagigue Creek
B - Conestogo River
C - Upper Grand River
D - Grand River, near drinking water intake
E - Grand River, near one wastewater treatment plant effluent point

3.2 Exposure Surveillance

Retail

As in previous years, a low prevalence of *Campylobacter* was detected on raw retail pork and beef (Table 3.1). Prevalence estimates were higher on retail poultry, consistent with previous surveillance years. When positive samples were enumerated, 75% of samples (69/92) were below the detection limit and 24% were found to have between 0.3 and 10 MPN/g (Appendix D).

New to 2009, bagged leafy greens (produce) were sampled at the retail level in the same manner as the raw meat sampling surveillance (Appendix C). Starting in April of 2009, 376 samples were collected and none were positive for *Campylobacter* spp. by culture.

Farm

Campylobacter coli was most commonly detected on swine farms (Table 3.1). *C. jejuni* was most commonly detected on dairy and beef farms. *Campylobacter* was not commonly detected on broiler chicken farms (5% of samples were positive).

To investigate the low prevalence observed in poultry droppings, parallel sampling with two laboratories and two methodologies was pursued, to improve *Campylobacter* recovery rates. In addition, transport media and time in transport have been investigated as possible explanations for the low prevalence. *Campylobacter* is notorious for its sensitivity to environmental changes and transition to a non-culturable state. We only sample flocks that are close to market and only collect fresh cecal droppings. This issue continues to be explored to develop a plausible hypothesis and inform future surveillance.

Water

Of the water samples that were positive for *Campylobacter*, half were *C. jejuni*. Surveillance data from 2009 exhibit similar trends to previous years (Table 3.1).

3.3   Temporal Distribution of Campylobacteriosis
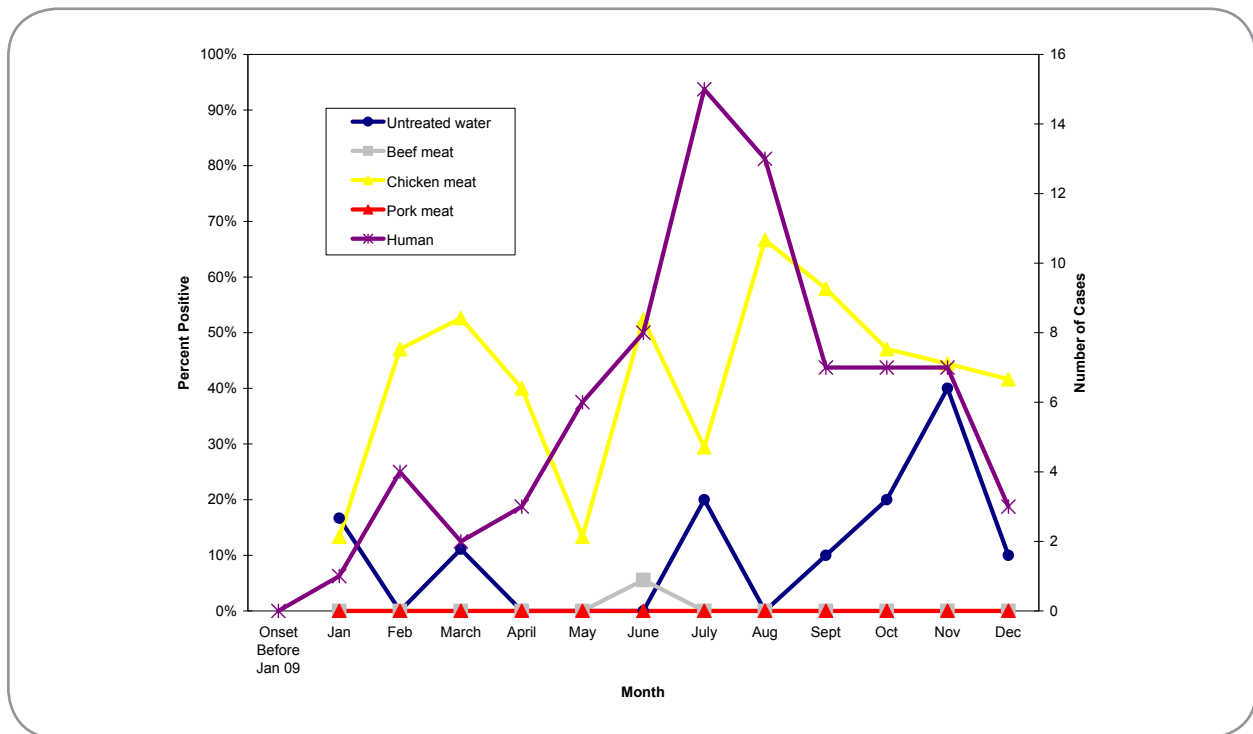
Figure 3.3. Monthly distribution of *Campylobacter jejuni* contamination from all sources and incidence of human cases in Sentinel Site 1 in 2009

3.4   Summary of *Campylobacter* Results

- Campylobacteriosis is the most frequently reported enteric disease in Sentinel Site 1.

- Contact with household pets and visiting a farm animal area continue to be a potential risk factor for *Campylobacter* infections in humans. Interestingly, the consumption of unpasteurized milk was not significant. However, consumption of raw milk still remains higher than the general (healthy) population. In previous years, this was identified as a potential risk factor for cases compared to all other cases (> 5% difference) (*2*).

- *C. jejuni* is the most common species associated with human campylobacteriosis.

- Raw chicken meat contaminated with *Campylobacter* carries a high proportion of *C. jejuni*. Pork and beef are rarely contaminated with *Campylobacter.*

- *Campylobacter* was not detected on bagged leafy greens.

- *C. coli* was detected on swine, beef and dairy farms, but not on poultry farms.

- *C. jejuni* and *C. lari* were detected in untreated surface water*; C. jejuni* was the most common species.

KEY MESSAGE:

Campylobacter jejuni is the most common species associated with human campylobacteriosis. *Campylobacter jejuni* is also the most commonly detected type of *Campylobacter* detected on raw, skinless chicken breasts purchased at retail. This highlights the importance of safe cooking and consumer handling practices.

4. *Salmonella*

4.1 Human Cases

In 2009, a total of 117 cases of salmonellosis were reported (22.6/100,000 person-years). Of these 117 cases, 30% (35) were travel-related (6.8/100,000 person-years) and 70% (82) were classified as endemic (15.8/100,000 person-years). There were no outbreak-related cases reported in 2009. In comparison, the annual incidence rates for salmonellosis in 2008 in Canada and Ontario were 18.2/100,000 and 18.9/100,000, respectively (*1*).

The age, gender and seasonal distributions fit patterns that have been historically observed for *Salmonella* (Figures 4.1 and 4.2). The infection is most common among children under ten years of age.
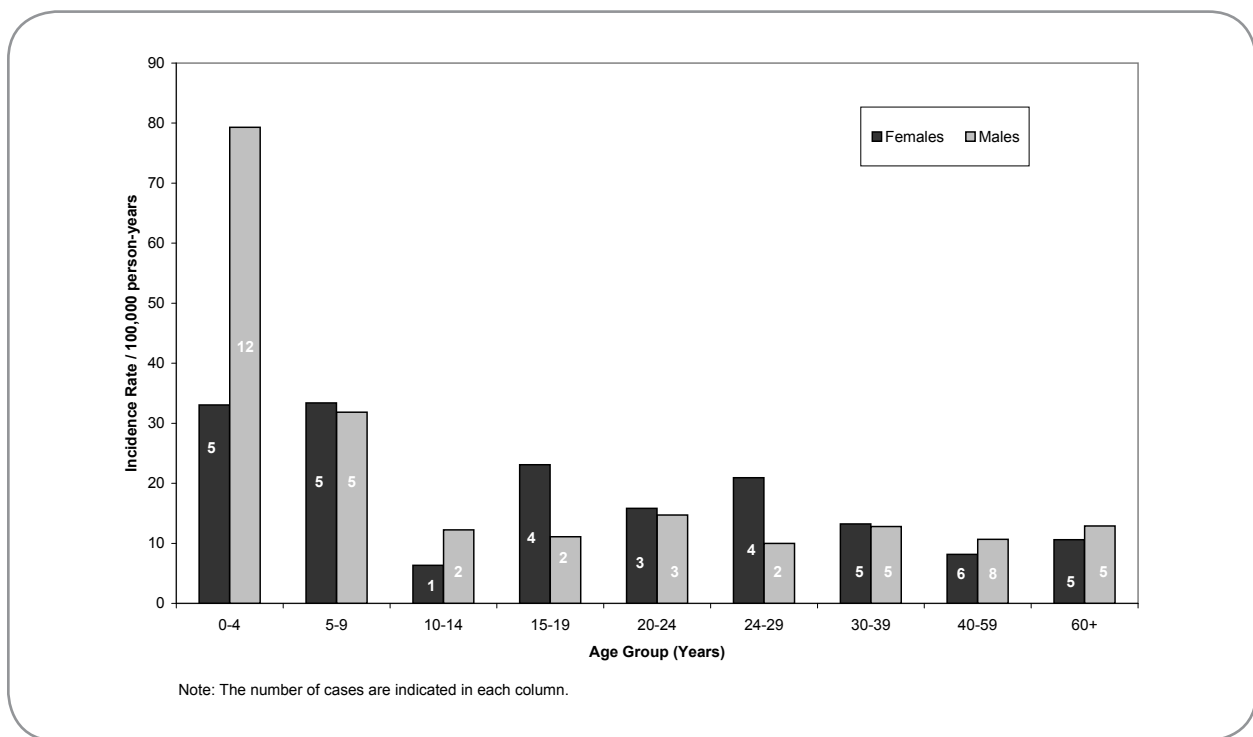
Figure 4.1. Incidence rates of endemic salmonellosis cases by gender and age group in Sentinel Site 1 in 2009

There were 21 different serotypes detected among the 82 endemic cases in 2009, for which the serotype was known. The top three serotypes, *S.* Enteritidis, *S.* Heidelberg, and *S.* Typhimurium comprised 64% (53/82) of isolates that were serotyped (Table 4.1).

Comparison of travel versus endemic *Salmonella* cases indicated that all (13/13) *S.* Heidelberg cases and 95% (20/21) of *S.* Typhimurium were of domestic origin. Conversely, 47% (17/36) of *S.* Enteritidis cases were travel-related.

Potential exposure information for the three days prior to onset of illness was collected for 91% (75/82) of the reported endemic *Salmonella* infections (Appendix B). Few meaningful risk factors were identified from the case-case comparison; however, eating in a restaurant (28%) and exposure to pet reptiles (9%) appeared to be specific risk factors for *Salmonella* cases.
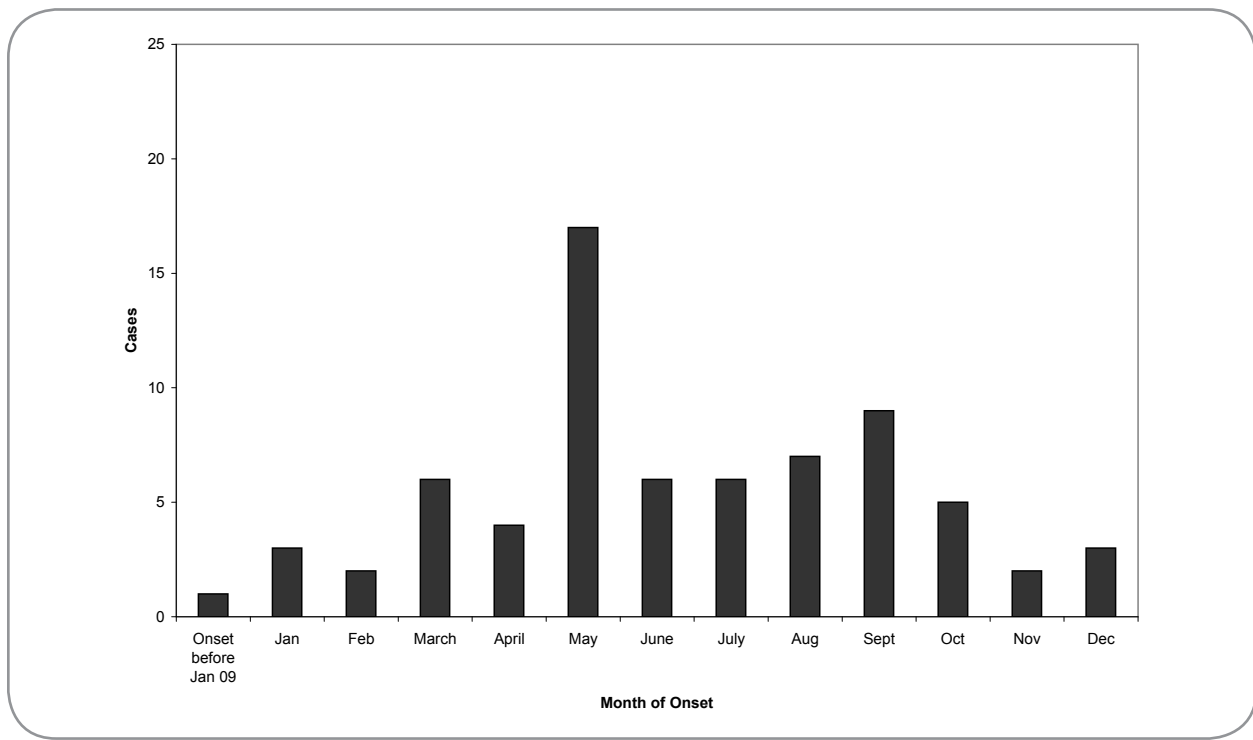
Figure 4.2. Temporal distribution of human endemic salmonellosis cases in Sentinel Site 1 reported in 2009

The incidence rate of endemic *Salmonella* was higher during the spring (March, April, May; Fisher's Exact Test: p=0.03), summer (June, July, August; p<0.05) and fall (September, October, November; p=0.01) than during the winter (December, January, February) (Figure 4.2).

4.2   Exposure Surveillance

Retail

Salmonella was detected in 29% (57/200) of retail skinless chicken breast samples collected in 2009 in Sentinel Site 1 (Table 4.1). It was rarely detected on retail pork chops (2% of samples) or ground beef (1% of samples). Based on the enumeration results, levels of cells detected on positive samples remained consistently low, as seen in previous surveillance years (Appendix D).

The three most frequent serotypes detected on retail chicken breasts were *S*. Kentucky, *S*. Heidelberg, and *S*. Enteritidis. Starting in April of 2009, 376 samples of bagged leafy greens (produce) were sampled at the retail level in the same manner as the raw meat sampling plan (Appendix C). None were positive for *Salmonella* spp. by culture.

On-Farm

In broiler chickens, the prevalence of *Salmonella* decreased significantly at the sample level from 62% (2008) to 31% (2009). At the farm level a decrease was also observed 76% (2008) to 53% (2009), but it was not significant (Table 4.1 and 2009 short report). Given that no changes were made in either the sample collection or testing methodologies this decrease might represent a trend towards reduced broiler flock *Salmonella* contamination in the sentinel site operations. Ongoing surveillance will illustrate whether this reduction remains steady from year to year.

Water

Salmonella was detected in 25% of surface water samples collected in 2009. Positive samples were collected at all 5 of the sample sites within the Grand River watershed.

TABLE 4.1
***Salmonella* detection and serotyping data (culture-based methods) from the integrated surveillance activities in Sentinel Site 1 in 2009.**

	Human	Retail Food				Food Animals (Manure)				Untreated Surface Water
	Endemic Cases	Pork	Chicken	Beef	Produce	Swine	Broiler Chickens	Beef Cattle	Dairy Cattle	Grand River
Detection		Pork chop	Chicken breast	Ground beef	Bagged leafy greens					5 sample points on Grand River
# tested	Unknown	200	200	200	376	120	120	120	120	112
# positive	82	3	57	1	0	41	37	15	22	28
% positive		2%	29%	1%	0%	34%	31%	13%	18%	25%
Serotyping[b]										
# serotyped	81	3	58	1		41	37	15	22	28
Agona						1				1 (B)
Albany			2							
Anatum	4									
Branderup	1					1				
Brandenburg						1				1 (C)
Cerro								5	11	1 (B)
Derby		1	1			7				2 (A)
Eastbourne	2									
Enteritidis	19		9				6			1 (E)
Hadar	1		4							
Hartford	3							1		
Heidelberg	14		15				4	2		
I:4,5,12:b:-	1									2 (C,E)
I:4,5,12:i:-	1					1				2 (D,E)
I:6,14,18:-:-									4	
Infantis			1			1			1	
Kentucky			22	1			22	2	5	3 (A,B,D)
London						3				
Mbandaka						1				
Muenchen	2									
Newport	2									3 (B,D,E)
Thompson	4		1							
Tyhpimurium	20	1	3			13	3	1	1	4 (A,C,D)
Uganda						1		2		
Worthington						7				1 (C)
Other[b]	8	1				4	2	1		8

Serotype ranking within each component
- most frequent serotype
- second most frequent serotype
- third most frequent serotype

[a] Includes var 5-.

[b] Serotypes that were identified once in a single component are listed below and are NOT listed in Table 4.1:

Human:	ssp. Arizonae (IIIA) 48:G,Z51:-, Havana, ssp. Houtenae (IV) 44:Z4,Z23:-, Saintpaul, Sandiego, ssp Enterica (I) OR:-:-, SSP I, Virchow
Retail Pork:	Adelaide
Swine Farms:	I:4,12:i:- , Johannesburg, Livingstone, Ohio, Stanley
Chicken Farms:	I:8,20:-z6, I:ROUGH-O:i:z6
Beef Farms:	I:ROUGH-O:fg:-
Untreated water:	I:6,7:-:1,5 (D), IIIb:11:k:- (C), IIIb:Rough-O:i:- (D), IV:50:-:- (D), Javiana (E), Poona (E), Tennessee (E)

Water Sampling Locations in Grand River Watershed:
A - Canagagigue Creek
B - Conestogo River
C - Upper Grand River
D - Grand River, near drinking water intake
E - Grand River, near one wastewater treatment plant effluent point

4.3 Advanced Subtype Comparison

One of the benefits of the C-EnterNet surveillance program is the application of enhanced laboratory methodologies to identify patterns in subtype distributions among both the human cases and potential sources over time. In this section, data on the top three serotypes associated with human salmonellosis infections in Canada (and in Sentinel Site 1) are more thoroughly presented, by phage type or PFGE pattern, and key trends are identified.

Salmonella Typhimurium

TABLE 4.2
Integrated comparison of *Salmonella* Typhimurium phage types for 2009
in Sentinel Site 1 (2005-2008 results in brackets)

Typhimurium	Human		Retail Food				Food Animals (Manure)				Untreated Surface Water
	Travel Cases	Endemic Cases	Pork	Chicken	Beef	Produce	Swine	Broiler Chickens	Beef Cattle	Dairy Cattle	Grand River
			Pork chop	Chicken breast	Ground beef	Bagged leafy greens					5 sample points on Grand River
# of samples with PT results	1 (2)	20 (48)	1 (3)	3 (12)			13 (51)	3 (1)	2(2)	1 (6)	4 (9)
PT1		(1)									
PT2		(1)					1 (1)				
PT3		1 (3)									
PT8		(1)									
PT10		(2)									
PT12/12A		1 (2)					2				
PT15							(1)				
PT21		(1)									
PT22		1									
PT28							(2)				
PT41		(1)									(1 D)
PT46							(1)				
PT51	1										
PT66		1									
PT69											(1 C)
PT82		(2)									
PT97		(2)									
PT104/104A		2 (4)	1 (1)	(3)			5 (25)	3	1(1)	1(5)	(2 A,C)
PT104B		1 (3)					(6)				
PT108	(1)	4 (16)		3 (2)				(1)			3 (C,D) (2 A,C)
PT110				(1)							
PT117		1									
PT120	(1)	(2)	(1)								
PT135				(2)							
PT151							(2)				
PT160									1		
PT169				(1)							
PT170		3 (1)		(1)							
PT193		1 (1)					(1)				(1 B)
PT194		1					(2)				
PT208		1		(1)			1 (2)				
PT1106		1									
PTU211a							2				1 (A)
PTU285		(1)									
PTU302		(1)	(1)				(6)		(1)	(1)	
PTU310		(1)									
PTUT1		(1)					2 (1)				
PTUT5							(1)				
Atypical		1 (1)		(1)							(2 A,B)

Water Sampling Locations in Grand River Watershed:
A - Canagagigue Creek
B - Conestogo River
C - Upper Grand River
D - Grand River, near drinking water intake
E - Grand River, near one wastewater treatment plant effluent point

S. Typhimurium was the most common serotype detected in swine manure, surface water, and is one of the top three serotypes among human cases in Sentinel Site 1 and nationally (PulseNet). As observed in Table 4.2, no clear associations are observed between human phage types and the potential sources. Phage type 104/104A was detected in the four commodity groups tested at the farm level in 2009 and on one retail pork sample and in two endemic human cases. Retail pork chops are typically negative for *Salmonella* (**5**). The most frequent subtype found in human cases (PT 108) was also the only one found in retail chickens, and was most frequent in untreated water.

Salmonella Enteritidis

TABLE 4.3
Integrated comparison of *Salmonella* Enteritidis phage types for 2009
in Sentinel Site 1 (2005-2008 results in brackets)

	Human		Retail Food				Food Animals (Manure)				Untreated Surface Water
	Travel Cases	Endemic Cases	Pork	Chicken	Beef	Produce	Swine	Broiler Chickens	Beef Cattle	Dairy Cattle	Grand River
Enteritidis			Pork chop	Chicken breast	Ground beef	Bagged leafy greens					5 sample points on Grand River
# of samples with PT results	17 (28)	18 (51)	(1)	9 (15)	(1)	0	0	6 (7)	(1)	0	1 (2)
PT1	2 (6)										
PT1A	(2)	(1)									
PT2				1				2			
PT4	1 (8)	(12)									
PT4A	(1)	(1)									
PT5B	6 (3)	(1)									
PT6		(1)									
PT6A	1 (3)	(1)									
PT6D				1							
PT8	1 (2)	6 (15)		5 (6)				1 (2)	(1)		
PT8A								(1)			
PT13		5 (12)	(1)	1 (7)	(1)			(3)			(2 A,B)
PT13A	3	6 (6)		1 (2)				2			
PT14B	1										
PT19								1			
PT21	1 (1)	(1)									
PT22											1 (E)
PT37	(1)										
PT911		(1)									
Atypical	1 (1)	1 (1)						(1)			

Water Sampling Locations in Grand River Watershed:
A - Canagagigue Creek
B - Conestogo River
C - Upper Grand River
D - Grand River, near drinking water intake
E - Grand River, near one wastewater treatment plant effluent point

Salmonella Enteritidis continues to increase in incidence in Canada, and has since 2005 (**1**). As noted in Table 4.3, *S.* Enteritidis is common among travel cases, yet particular phage types are most common among endemic cases, including PT8, PT13, and PT13A. One of the main sources of *S.* Enteritidis is believed to be poultry products, including eggs and chicken meat (**3**). As noted by these surveillance data from 2009, these three phage types of *S.* Enteritidis were detected on retail chicken, in broiler chicken manure samples, and in human domestic cases, supporting this hypothesis.

Salmonella Heidelberg

S. Heidelberg data are presented by phage type (Table 4.4) and PFGE (Table 4.5), to illustrate the different utilities of these molecular typing methods. It is clear that for the exposure source monitoring, by either method, *S*. Heidelberg is most commonly identified on retail chicken breasts, and on the broiler chicken manure farms. There is a broad distribution of phage types (or PFGE patterns) detected on retail chicken meat.

By phage type, there is some alignment between human endemic cases, retail chicken meat, broiler chicken manure and untreated surface water for PT19, and for PT29 to a smaller extent (Table 4.4). Conversely, this overlap is not observed as clearly by PFGE (Table 4.5), since all 4 isolates detected on broiler chicken farms from 2009 were identified as one PFGE pattern (SHEXAI.0001). This particular PFGE pattern is common in human cases, both in Sentinel Site 1 and nationally (**4**).

TABLE 4.4
Integrated comparison of *Salmonella* Heidelberg phage types for 2009
in Sentinel Site 1 (2005-2008 results in brackets)

	Human		Retail Food				Food Animals (Manure)				Untreated Surface Water
	Travel Cases	Endemic Cases	Pork	Chicken	Beef	Produce	Swine	Broiler Chickens	Beef Cattle	Dairy Cattle	Grand River
Heidelberg			Pork chop	Chicken breast	Ground beef	Bagged leafy greens					5 sample points on Grand River
# samples with Phagetype results	0 (0)	12 (7)	0	15 (40)	0	0	0	4 (6)	2	0	0 (2)
PT2		2		(3)							
PT4									1		
PT5				1 (6)							
PT9				(4)							
PT10				1 (1)							
PT11		(1)		(1)							
PT11a				(2)							(1 A)
PT17				1				(1)			
PT18		1 (1)		1							
PT19		8 (1)		6 (14)				1	1		(1 C)
PT19a				(1)							
PT25				(1)							
PT26				(1)							
PT29		(1)		4 (1)				2			
PT35				1							
PT39				(1)							
PT41		1 (2)		(1)							
PT46								(5)			
PT52				(1)							
Atypical		(1)		(2)				1			

Water Sampling Locations in Grand River Watershed:
A - Canagagigue Creek
B - Conestogo River
C - Upper Grand River
D - Grand River, near drinking water intake
E - Grand River, near one wastewater treatment plant effluent point

	Human		Retail Food			Food Animals (Manure)				Untreated Surface Water
	Travel Cases	Endemic Cases	Pork	Chicken	Beef	Swine	Broiler Chickens	Beef Cattle	Dairy Cattle	Grand River
			Pork chop	Chicken breast	Ground beef					5 sample points on Grand River
Heidelberg										
# samples with PFGE results	0	13 (8)	0	15 (41)	0	0	4 (6)	2 (0)	0	0 (2)
SHEXAI.0001		3 (1)		10 (16)			4 (6)	1		(1)
SHEXAI.0006		(1)		(5)						(1)
SHEXAI.0007		8		(3)				1		
SHEXAI.0009		(4)		1						
SHEXAI.0011		(2)		2 (6)						
SHEXAI.0015				(1)						
SHEXAI.0020				2 (7)						
SHEXAI.0187				(1)						
SHEXAI.0194		1		(1)						
SHEXAI.0201		1								
SHEXAI.0204				(1)						

Water Sampling Locations in Grand River Watershed:
A - Canagagigue Creek
B - Conestogo River
C - Upper Grand River
D - Grand River, near drinking water intake
E - Grand River, near one wastewater treatment plant effluent point

Other Serotypes

S. Kentucky is commonly found on retail chicken (33% of positive samples) and in the broiler chicken manure samples (59% of positive samples), and occasionally detected in the surface water, but it is not commonly detected in humans (no cases were detected in Sentinel Site 1 in 2009) (Table 4.1). This trend has been repeated year to year in Sentinel Site 1 (**5**). The epidemiology of *S.* Kentucky is important to monitor, since the surveillance data suggest that it is prevalent in a number of potential sources, yet does not contribute to the human burden of salmonellosis. Future surveillance and research will help to understand whether this trend will continue, and which factors are influencing the limited cases in people.

In 2009, *S.* Cerro was most commonly detected in both beef and dairy manure samples, yet was not isolated in any human case in Sentinel Site 1, nor is it commonly seen nationally (**6**).

4.4   Temporal Distribution
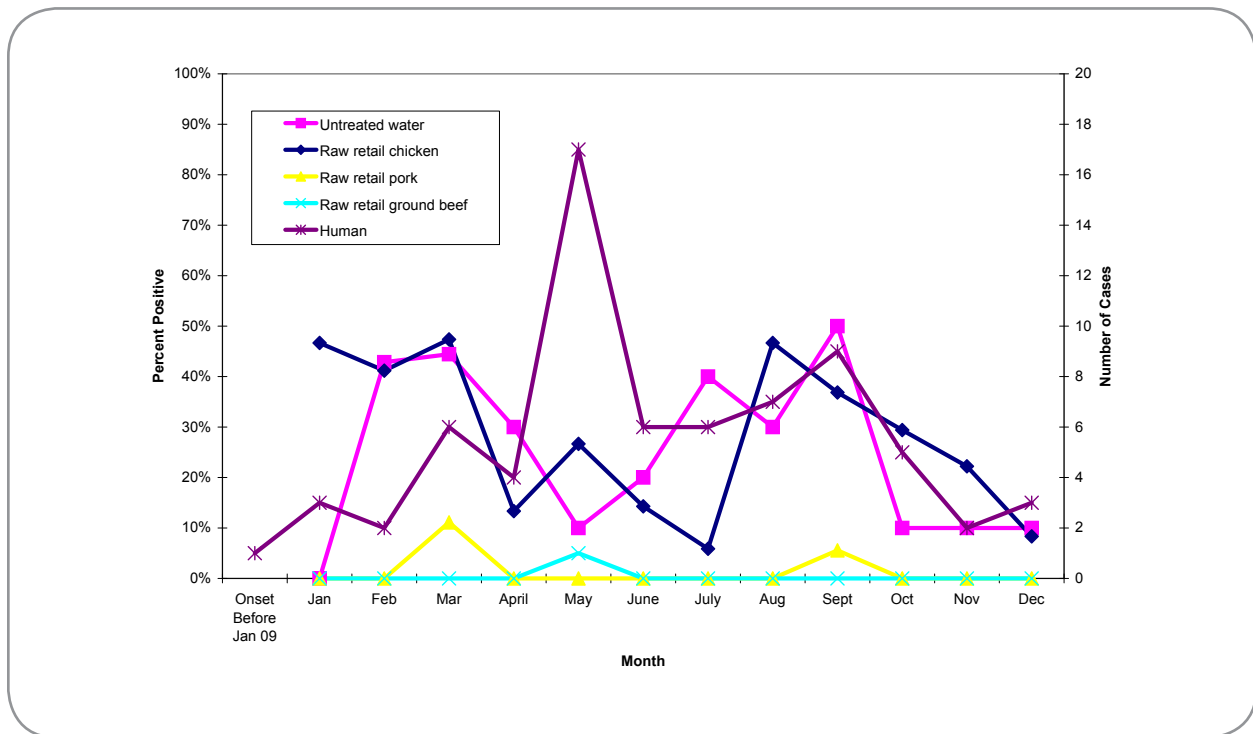
Figure 4.3. Temporal distribution of *Salmonella* detected in human endemic cases, untreated surface water and retail meat samples in Sentinel Site 1 in 2009

No obvious seasonal patterns were observed in the detection of *Salmonella* in the exposure sources within Sentinel Site 1 in 2009. Seventeen of the human cases were reported in May (including *S*. Anatum, Branderup, Eastbourne, Enteritidis, Heidelberg, Thompson and Typhimurium), suggesting a late spring peak, and a range of 1-9 cases was observed throughout the rest of the year.

4.5   Summary of *Salmonella* Results

- The human salmonellosis burden in Sentinel Site 1 is mostly associated with *S*. Typhimurium, *S*. Enteritidis and *S*. Heidelberg.

- In 2009, the peak in endemic cases was observed in May.

- One of the main sources of *S*. Enteritidis infection is believed to be poultry products, including eggs and chicken meat (**3**). The 2009 surveillance data illustrate that the three phage types of *S*. Enteritidis associated with domestic infections (PT8, PT13 and PT13A) were detected on retail chicken and broiler chicken manure samples, also supporting this hypothesis.

- *S*. Heidelberg is a domestically-acquired infection, and exposure source monitoring illustrates alignment for some phage types (PT19) among human cases and detection on retail chicken meat and on-farm broiler chicken manure and untreated surface water sampling.

KEY MESSAGE:

Enhanced, standardized laboratory testing across all C-EnterNet components (human, farm, retail and water) has allowed for the identification of patterns in subtype distributions among human cases and potential exposure sources over time.

Possible associations have been observed between human cases of *S*. Enteritidis and poultry, particularly for PTs 8, 13 and 13A. Other serotypes (*S*. Kentucky and *S*. Cerro) are commonly detected among exposure sources but are rarely detected in humans. Further surveillance will continue to inform prevention and control efforts.

5. Pathogenic *E. coli*

5.1 Human Cases

In 2009, in Sentinel Site 1, there were 10 reported cases of verotoxigenic *E. coli* infections, 9 of which were *E. coli* O157:H7 and 1 *E. coli* O103:H29. All 10 cases were classified as endemic (1.9/100,000 person-years). No travel-related cases were reported. In comparison, the annual incidence rates for *E. coli* O157:H7 in 2008 in Canada and Ontario were 2.3/100,000 and 2.2/100,000, respectively (*1*).

The age- and gender-specific incidence rates among the 10 endemic cases shows that females between 30-39 had the highest rates (7.9/100,000 person-years), followed closely by children less than 5 years of age (Figure 5.1).

Note: The number of cases are indicated in each column.
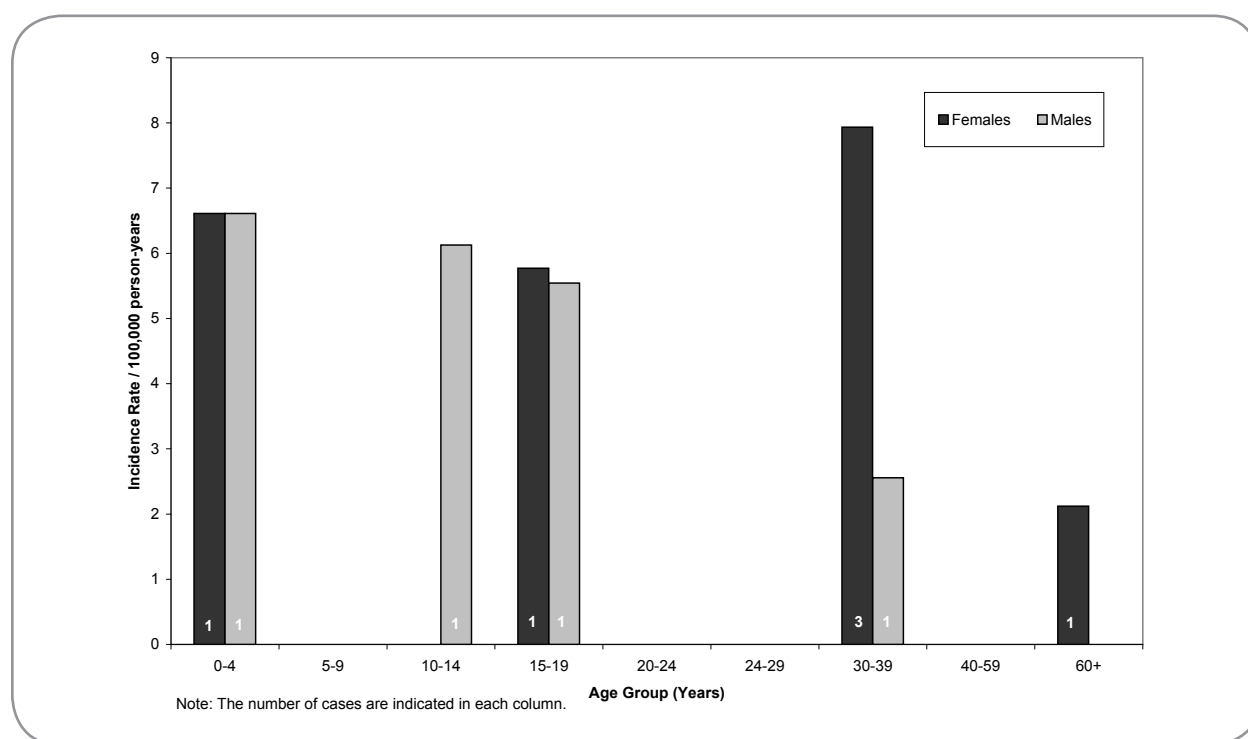
Figure 5.1. Incidence rates of endemic *E. coli* O157:H7 in Sentinel Site 1 by gender and age group in 2009

Risk factor information for the ten days prior to the onset of illness was collected for 100% (10/10) of the reported endemic cases of *E. coli* O157:H7 (Appendix B). A higher number of *E. coli* O157:H7 cases was observed for the following factors: using a private well; drinking unpasteurized milk; attending a barbeque; eating in a restaurant; eating meat from a butcher shop; eating meat from a private kill; shopping at a butcher shop; having contact with household dogs; visiting a farm animal area (with cattle and pigs); and living on a farm and had on-farm animal contact with pigs and sheep. Other risk factors observed among *E. coli* O157:H7 cases included travel within Canada by car ($P \leq 0.05$), and canoeing, kayaking, hiking or camping ($P \leq 0.05$).

5.2 Exposure Surveillance

TABLE 5.1
Verotoxigenic *E. coli* detection data from the
integrated surveillance activities in Sentinel Site 1 in 2009.

	Human	Retail Food				Food Animals (Manure)				Untreated Surface Water
	Endemic Cases	Pork	Chicken	Beef	Produce	Swine	Broiler Chickens	Beef Cattle	Dairy Cattle	Grand River
Detection		Pork chop	Skin-off breast	Ground beef	Bagged Leafy Greens	30 Farms	30 Farms	30 Farms	30 Farms	5 sample points on Grand River
# tested	Unknown	200	200	200	376	120	120	120	120	112
# positive	10	0	0	1	0	7 (4 farms)	0 (0 farms)	12 (6 farms)	7 (5 farms)	0
Percentage positive (%)		0%	0%	1%	0%	6%	0%	10%	6%	0%
VTEC		0	0	1						
O103:H29	1									
O157 (non-H7)						3		1		
O157:H7	9					4		11	7	

Retail

Verotoxigenic *E. coli* (VTEC) was detected on 1% of retail beef samples (Table 5.1). VTEC was not detected on retail pork or chicken samples. New in 2009, bagged lettuce was sampled for VTEC by molecular PCR, and all samples were negative.

On-Farm

E. coli O157:H7 was isolated from 9% (11/120) of the pooled manure samples collected from beef operations and from 6% (7/120) of the pooled manure samples collected from dairy operations (Table 5.1). None of the broiler chicken manure samples tested positive for *E. coli* O157:H7. *E. coli* O157:H7 was also isolated from 3% (4/120) of swine manure samples, collected from two swine farms. However, on one of those farms there were beef cattle, and on the other, the strains were considered non-pathogenic, since they lacked the shiga toxins 1 and 2.

Water

E. coli O157:H7 was not detected in any of the 112 water samples collected along the Grand River in 2009.

TABLE 5.2
PFGE results for *E. coli* O157:H7 for all components, including human travel-related cases in Sentinel Site 1 in 2009 (values in brackets refer to results from 2005-2008 for comparison)

	Human		Food Animals (Manure)			Untreated Surface Water
	Non-travel Cases	Travel-related Cases	Swine	Beef Cattle	Dairy Cattle	Grand River
						5 sample points on Grand River
# of isolates with PFGE results	7(61)[a]	0 (3)	4 (1)	11 (21)	7 (42)	0 (7)
ECXAI.0001	(5)			(2)	(1)	
ECXAI.0002	(1)					
ECXAI.0006				(3)	(3)	
ECXAI.0007	(1)					
ECXAI.0008	1(2)			(1)	(1)	(1)
ECXAI.0012				1		
ECXAI.0017	(3)					
ECXAI.0023					(1)	
ECXAI.0052	1(2)	(1)				
ECXAI.0063	(1)					
ECXAI.0073				(1)		
ECXAI.0096	(1)					
ECXAI.0140				(1)		
ECXAI.0221	1					
ECXAI.0247	(1)					
ECXAI.0262	(9)					
ECXAI.0266				2		
ECXAI.0309	(1)					
ECXAI.0317					(1)	
ECXAI.0378					(1)	
ECXAI.0407				(2)		
ECXAI.0776				(1)		
ECXAI.0825				2 (1)		
ECXAI.0841	(1)					
ECXAI.1164				(1)		
ECXAI.1175	(1)				1 (1)	
ECXAI.1182					1	
ECXAI.1186	1					
ECXAI.1216			1			
ECXAI.1221	(1)					
ECXAI.1248	(1)					
ECXAI.1267				(1)	(1)	
ECXAI.1301	1					
ECXAI.1304					(1)	
ECXAI.1325					1	
ECXAI.1456	1					
ECXAI.1477	(1)					
ECXAI.1478	(1)					
ECXAI.1495	(1)					
ECXAI.1501	(1)					
ECXAI.1526	(1)					
ECXAI.1537	(1)					
ECXAI.1556						(4)
ECXAI.1557						(1)
ECXAI.1577	(2)					
ECXAI.1578	(1)					
ECXAI.1599					1	
ECXAI.1610	(1)					
ECXAI.1611					(3)	
ECXAI.1612					(3)	
ECXAI.1613					(2)	
ECXAI.1614					(1)	
ECXAI.1687					(6)	
ECXAI.1688					(1)	
ECXAI.1689					(1)	
ECXAI.1690					(4)	
ECXAI.1691					(1)	
ECXAI.1692	(1)				(2)	
ECXAI.1694	(1)				(2)	
ECXAI.1714		(1)				
ECXAI.1737	(2)					
ECXAI.1777		(1)				
ECXAI.1844						(1)
ECXAI.1855					(1)	
ECXAI.1857					(1)	
ECXAI.1858				(1)		
ECXAI.1859				(1)		
ECXAI.1860				(1)		
ECXAI.1898	(2)					
ECXAI.1901	(1)					
ECXAI.1940	(1)					
ECXAI.1972	(1)					
ECXAI.2003					(1)	
ECXAI.2108				(1)		
ECXAI.2109					(1)	
ECXAI.2110				(2)		
ECXAI.2111			(1)			
ECXAI.2112					(1)	
ECXAI.2172				(1)		
ECXAI.2239	1					
ECXAI.2324			1			
ECXAI.2325			1			
ECXAI.2327				1		
ECXAI.2328					1	
ECXAI.2329					1	
ECXAI.2330				2		
ECXAI.2378					1	
ECXAI.2379				1		
ECXAI.2380				1		
ECXAI.2381			1			
ECXAI.2382				1		

[a] Non-travel includes endemic and outbreak cases.

PFGE analysis of the 2009 *E. coli* O157:H7 isolates showed 29 isolates comprising 25 distinct PFGE patterns and no overlap between human cases and isolates from non-human sources (Table 5.2). One case of ECXAI.0008 was detected, which is the third most common pattern in the PulseNet Canada database (associated with 16 human cases in 2009). Interestingly, the most frequently occurring PFGE pattern among human clinical isolates reported to PulseNet Canada for 2009, ECXAI.0001, was not recovered from any of the C-EnterNet surveillance components in 2009.

When comparing five years of surveillance data, very little overlap was found among PFGE patterns. Some overlap was observed between dairy and beef cattle isolates, and in one year, one of these PFGE patterns was also detected in a surface water sample. There is considerable diversity in *E. coli* O157:H7 PFGE patterns, as observed both nationally and within the C-EnterNet program, so these results are not surprising.

5.3   Temporal Distribution

Endemic VTEC cases were reported between March and November. The highest number of cases (3 per month) was reported in August and September.

5.4   Summary of Pathogenic *E. coli* Results

- *E. coli* O157:H7 continues to be a domestically acquired infection, as demonstrated by the absence of travel-related cases in 2009.

- PFGE subtyping of the human and non-human isolates from 2009 revealed no overlapping patterns, suggesting that different strains are circulating in these components. When reviewing data from multiple years, little overlap exists.

- Travel within Canada by car ($P{\leq}0.05$), and canoeing, kayaking, hiking or camping ($P{\leq}0.05$) may increase the risk for this domestically-acquired infection.

KEY MESSAGE:

Very little overlap is observed between PFGE patterns of human *E. coli* cases and *E. coli* isolates detected in the exposure sources. Continued enhanced, integrated surveillance and targeted studies to better understand the potential sources of this domestically acquired infection are needed.

6. *Yersinia*

6.1   Human Cases

In 2009 in Sentinel Site 1, there were 8 reported cases of *Yersinia* infection (1.5/100,000 person-years). Of these 8 cases, 13% (1) were travel-related (0.20/100, 000 person-years), and 87% (7) were classified as endemic (1.4/100,000 person-years). Currently, *Yersinia* is not a nationally-notifiable disease, and so the annual national and provincial incidence rates are not available for comparison. The age-specific incidence rate from the 7 endemic cases was highest among male children less than five years of age (19.8/100,000 person-years). (Figure 6.1).
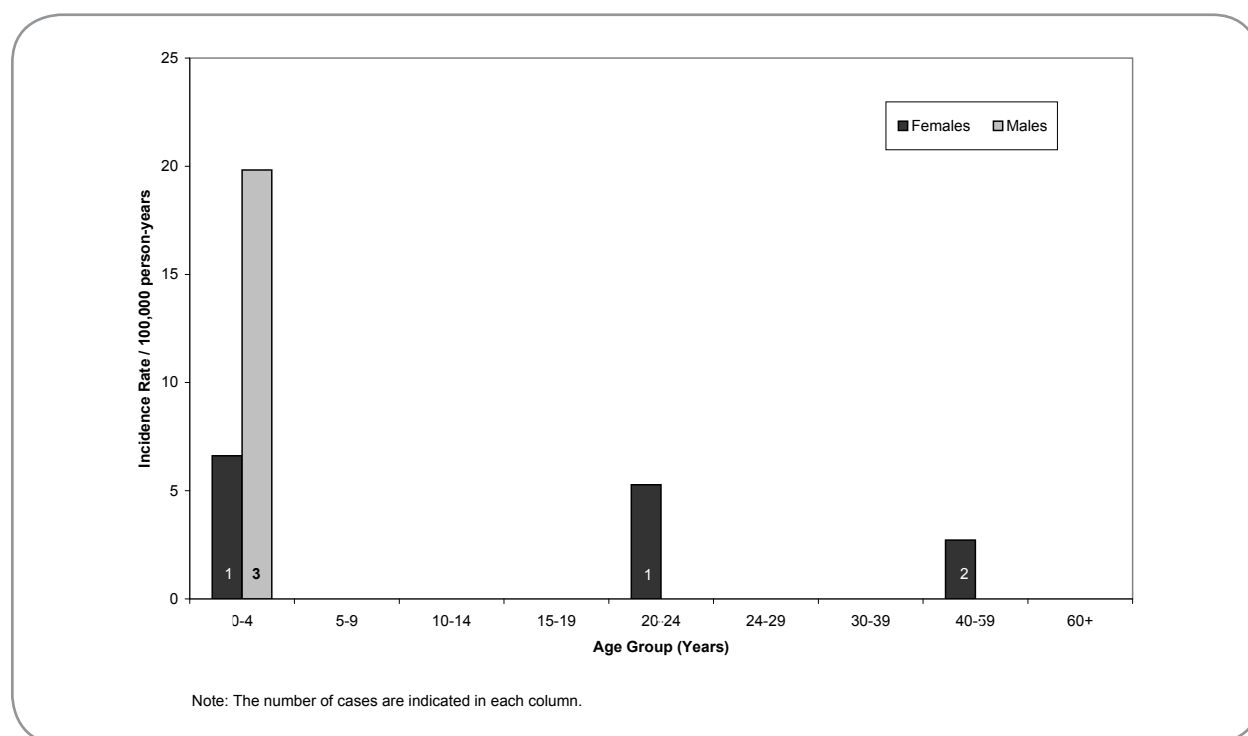
Figure 6.1. Incidence rates of endemic *Yersinia* infection by gender and age group in Sentinel Site 1 in 2009

Of the human *Yersinia* infections, 6/6 were subtyped as *Y. enterocolitica* biotype 4, serotype O:3-, considered to be a pathogenic strain. The cases were uniformly spread over the year without obvious seasonal patterns.

Potential exposure information for the seven days prior to the onset of illness was collected for 86% (6/7) of the reported endemic yersiniosis cases (Appendix B). A higher number of reported yersiniosis cases were observed for the following exposures: swimming in a pool, eating meat from a butcher shop, and shopping at a butcher shop.

6.2   Exposure Surveillance

TABLE 6.1
***Yersinia* detection and speciation data from the integrated
surveillance activities in Sentinel Site 1 in 2009**

	Human	Retail Food	Food Animals (Manure)	Untreated Surface Water
	Endemic Cases	Pork	Swine	Grand River
Detection		Pork chop	30 farms	5 sample points on Grand River
# tested	Unknown	200	120	112
# positive	7	60	2 (2 farms)	49
% positive		30%	2%	44%
Subtyping				
# subtyped	6	58ᵃ		49ᵃ
Y. aldovae - non-pathogenic				2 (A,B)
Y. bercovieri - non-pathogenic		1		2 (B, E)
Y. enterocolitica - pathogenic	6	1	2	
Y. enterocolitica - non-pathogenic		32		13 (A,B,C,D,E)
Y. frederiksenii - non-pathogenic		12		7 (A,B,D,E)
Y. intermedia - non-pathogenic		12		20 (A,B,C,D,E)
Y. kristensenii - non-pathogenic		1		2 (A,C)
Y. mollaretti - non-pathogenic				3 (A,B,C)
Y. rohdei - non-pathogenic				

ᵃ Multiple isolates were detected in more than one samples, 59 isolates in total

Water Sampling Locations in Grand River Watershed:
A - Canagagigue Creek
B - Conestogo River
C - Upper Grand River
D - Grand River, near drinking water intake
E - Grand River, near one wastewater treatment plant effluent point

Retail

Yersinia was isolated from 30% (60/120) of the raw pork chops sampled (Table 6.1). This increase in prevalence is likely due to the method change that was implemented during the summer of 2009 to increase sensitivity (Appendix C). With the increase in samples positive for *Yersinia*, more samples (31%) were found to have measurable levels of cells (above the MPN detection limit) (Appendix D).

Of the 60 positive samples, only 1 was pathogenic (biotype 4, serotype O:3). All of the other *Yersinia* detected on retail pork samples were non-pathogenic. The non-pathogenic isolates were biotype 1A, serotypes O:5; O:7,13; O: Rough; and O:untypable.

On-Farm

Yersinia was isolated from 2% (2/120) of the pooled swine manure samples collected on the 30 farms (Table 6.1). Both isolates were pathogenic *Y. enterocolitica* (biotype 4, serotype O:3).

Water

All *Y. enterocolitica* isolates (O:5; O:7,13; O:7,8; O:Rough) from the untreated surface water samples were non-pathogenic.

6.3 Summary of *Yersinia* Results

- Based on this year and previous year's data, *Yersinia* continues to be a domestically-acquired infection, as demonstrated by the low proportion of travel-related cases.

- Epidemiologically, swimming in a pool and meat from a butcher shop may be important risk factors for yersiniosis.

- Pathogenic (biotype 4, serotype O:3:-) *Yersinia enterocolitica* were identified in pooled swine manure and on 1 retail pork sample, for the first time in 2009.

- All *Yersinia* detected in untreated surface water samples were non-pathogenic.

KEY MESSAGE:

The incidence rate of yersiniosis is relatively lower than other enteric infections. However, it appears to be mostly a domestically-acquired infection with potential association to swine (both manure and retail pork meat, though only one human-pathogenic strain was isolated on pork chops in five years of surveillance).

7. *Listeria*

7.1   Human Cases

Human listeriosis is rare and is typically identified with severe, hospitalized cases among immunocompromised individuals. An annual national incidence rate for listeriosis is not currently available from the NND. Health Canada's *Listeria* Reference Services, however, reports the incidence remains below 0.72 cases per 100,000 person-years nationally (*7*). One endemic case was detected in 2009 in Sentinel Site 1.

7.2   Exposure Surveillance

In 2009, *Listeria monocytogenes* testing was not continuous. For the retail component, testing was initiated in March 2009 on retail raw meats and produce (ie: bagged leafy greens). Testing was discontinued in 2008 within the farm component.

TABLE 7.1
***Listeria monocytogenes* detection data from the integrated**
surveillance activities in Sentinel Site 1 in 2009

	Human		Retail Food			
	Endemic	Outbreak	Pork[a]	Chicken[a]	Beef[a]	Produce[a]
Detection			Pork Chop	Skin-off breast	Ground beef	Bagged Leafy Greens
# samples tested	Unknown	Unknown	163	165	164	376[b]
# positive	1	0	16	28	20	5
% positive			10%	17%	12%	1%

[a] Sampled between March and December

[b] Tested by culture method

Retail

Given the lack of continuous testing for *Listeria monocytogenes* on retail meats between years, no direct comparisons amongst surveillance years are made (Table 7.1). In 2009, bagged leafy greens were added to the sampling protocol. Three samples were positive for *Listeria monocytogenes* in 2009 (culture-based testing). Enumeration results indicated that the majority of positive samples (69% of pork, 75% of chicken, and 75% of beef) were below the enumeration method detection limit (Appendix D).

TABLE 7.2
***Listeria monocytogenes* serotype data from the integrated surveillance activities in Sentinel Site 1 in 2009 (values in brackets refer to 2005-2008 data for comparisons)**

Serotype	Human		Retail Food				Farm Animals (manure)				Non-Human Total
			Pork	Chicken	Beef	Produce	Swine	Broiler Chickens	Beef Cattle	Dairy Cattle	
	Endemic	Outbreak	Pork Chop	Skin-off breast	Ground beef	Bagged leafy greens					
# serotyped	1 (3)	0 (3)	16 (43)	28 (136)	20 (107)	5	Not Tested (4)	Not Tested (8)	Not Tested (74)	Not Tested (15)	67
1/2a	1 (2)	(3)	5 (18)	17 (92)	10 (45)	3	(1)	(5)	(33)	(2)	34
1/2b			4 (13)	4 (27)	9 (57)		(3)	(3)	(12)	(4)	17
1/2c			7 (10)	2 (6)	1 (4)						
3a			(1)	(2)	(1)						10
3b				1 (5)							1
4a									(4)		
4b	(1)		(1)	4 (4)		2			(21)	(5)	5
4c									(4)	(4)	

Subtype Comparisons

Listeria monocytogenes serotypes 1/2a, 1/2b and 4b are frequently detected in the exposure sources tested and are reported to be the predominant serotypes in Canada that cause human illness (**8**). In this sentinel site, the one human *Listeria monocytogenes* isolate was serotyped as 1/2a, showing overlap between human cases and exposure sources. Of the top three human serotypes detected nationally, 1/2a was detected on all retail meats and produce and 4b was detected on chicken meat and produce (Table 7.2).

When comparing PFGE patterns from human and retail meat and produce samples collected in 2009, no predominant subtype emerges across species and sampling levels, (Table 7.3). The one human case identified in 2009 had PFGE pattern LMAAI.0003, which has historically been detected on retail meats in Sentinel Site 1 (Table 7.3). PulseNet data were used to identify the most common human PFGE patterns in 2009 on a national level, to compare the sentinel site data with Canadian data. The three most common PFGE patterns reported to PulseNet in 2009 were LMAAI.0003 (5 human cases, and in previous years detected on pork, beef and chicken meat), LMAAI.0287 (5 human cases and detected on chicken meat), and LMAAI.0234 (4 human cases and detected on one sample of bagged leafy greens). These case numbers are significantly lower than observed in 2008.

TABLE 7.3

***Listeria monocytogenes* PFGE data from the integrated surveillance activities in Sentinel Site 1 in 2009 (values in brackets refer to 2005-2008 data for comparison).**

PFGE Pattern	Human	Retail Food				Farm Animals (Manure)				Non-human Total
	Endemic Cases	Pork	Chicken	Beef	Produce	Swine	Broiler Chickens	Beef Cattle	Dairy Cattle	
		Pork Chop	Skin-off breast	Ground beef	Bagged leafy greens					
Number subtyped	1 (3)	16 (43)	28 (136)	20 (107)	5	Not Tested (4)	Not Tested (8)	Not Tested (74)	Not Tested (15)	
LMAAI.0001	(1)	(3)	(17)	1 (5)	1					1 (25)
LMAAI.0003	1	(1)	(1)	(1)						(3)
LMAAI.0007								(3)		(3)
LMAAI.0013		2 (8)	3 (24)	5 (23)						10 (55)
LMAAI.0014		(1)								(1)
LMAAI.0015			1							
LMAAI.0017								(1)		(1)
LMAAI.0024		(2)	2 (1)	(4)						2 (7)
LMAAI.0028		2	(5)	1 (1)						3 (6)
LMAAI.0049			(2)	(1)				(2)		(5)
LMAAI.0074			(3)					(2)	(1)	(6)
LMAAI.0090								(1)	(1)	(2)
LMAAI.0093	(1)			(1)			(1)	(11)		(13)
LMAAI.0096					2					2
LMAAI.0097			1 (9)							1 (9)
LMAAI.0126		2	(4)	2 (3)				(5)[b]		4 (12)
LMAAI.0147		4 (2)								4 (2)
LMAAI.0149										
LMAAI.0165		1								1
LMAAI.0193			5 (1)					(1)		5 (2)
LMAAI.0204								(9)[c]	(5)	(14)
LMAAI.0223		(9)	(2)	(45)						(56)
LMAAI.0234					1					1
LMAAI.0256		(1)		(1)						(2)
LMAAI.0265	(1)									
LMAAI.0266								(5)		(5)
LMAAI.0287			2							2
LMAAI.0317								(1)		(1)
LMAAI.0333								(1)	(1)	(2)
LMAAI.0352			1							1
LMAAI.0360			(2)							(2)
LMAAI.0377			(3)							(3)
LMAAI.0378		(5)		(2)						(7)
LMAAI.0381			(2)							(2)
LMAAI.0382				1 (1)						1 (1)
LMAAI.0383			2 (2)							2 (2)
LMAAI.0384		1 (1)		(1)						1 (2)
LMAAI.0392			1				(1)			1 (1)
LMAAI.0402			1 (10)							1 (10)
LMAAI.0403							(1)			(1)
LMAAI.0404							(1)			(1)
LMAAI.0407								(1)		(1)
LMAAI.0409								(1)		(1)
LMAAI.0411		1	(1)					(1)		1 (2)
LMAAI.0413								(1)		(1)
LMAAI.0414								(1)		(1)
LMAAI.0415								(1)		(1)
LMAAI.0418								(1)		(1)
LMAAI.0420								(1)		(1)
LMAAI.0421								(1)		(1)
LMAAI.0423		(1)						(1)		(2)
LMAAI.0424								(1)		(1)
LMAAI.0425								(1)		(1)
LMAAI.0427							(1)			(1)
LMAAI.0428							(1)			(1)
LMAAI.0429								(1)		(1)
LMAAI.0430								(1)		(1)
LMAAI.0431								(1)		(1)
LMAAI.0432						(2)				(2)
LMAAI.0433			(1)				(1)			(2)
LMAAI.0442			1							1
LMAAI.0454			(3)							(3)
LMAAI.0455			1 (2)							1 (2)
LMAAI.0465			(7)							(7)
LMAAI.0467			1 (2)	(1)						1 (3)
LMAAI.0472			(2)							(2)
LMAAI.0482			1							1
LMAAI.0483			1							1
LMAAI.0498			(2)							(2)
LMAAI.0531			(2)							(2)
LMAAI.0565		2 (1)	1	8 (4)						11 (5)
LMAAI.0584			1							1
LMAAI.0654				(1)						(1)
LMAAI.0851			1							1
LMAAI.0852		1								1
LMAAI.0855				1						1
LMAAI.0864			1							1
LMAAI.0880				1						1
Other patterns[a]		(8)	(23)	(12)	1	(2)		(11)	(4)	(60)
No PFGE designation			(3)				(1)	(6)	(3)	(13)

[a] PFGE patterns that were identified once in a single component

[b] 2 isolates found on the same farm

[c] 3 isolates found on the same farm

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7.3 Summary of *Listeria monocytogenes* Results

- As in previous years, pathogenic strains of *Listeria monocytogenes* were found on retail skinless chicken breast, pork chops and ground beef.

- Literature suggests that abattoirs and meat processing environments rather than farm animals may be an important source of *Listeria monocytogenes* (**9**), and while farms were not sampled in 2009, the retail meat data illustrate the presence of pathogenic serotypes on chicken, beef, pork and bagged leafy greens.

- When comparing the 2009 human endemic case results, with PFGE pattern LMAAI.0003 and serotype 1/2a, to historical exposure sources, there is overlap with retail pork, chicken, and ground beef.

- When comparing historical human endemic case results to exposure sources, there is overlap, with PFGE pattern LMAAI.0093 (human, beef, broiler chickens, and beef cattle) and with serotypes 1/2a and 4b (retail meat and farm manure).

KEY MESSAGE:

Listeria monocytogenes appears to be common in various retail food products, including raw meat and bagged leafy greens, and is also detected on the farm. Continuous surveillance will help identify potential sources of exposure to *Listeria monocytogenes*. Identifying the impact of this infection in vulnerable populations may further inform prevention and control efforts.

8. Parasites

8.1 *Giardia*

In 2009, there were a total of 72 reported cases of giardiasis (13.9/100,000 person-years). Of these 72 cases, 40 (56%) were endemic (7.7/100,000 person-years) and 32 were classified as travel-related (6.2/100,000 person-years). There were no outbreak-related cases. In comparison, the annual incidence rates for giardiasis in 2008 in Canada and Ontario were 12.7/100,000 and 12.4/100,000, respectively (*1*). Of the endemic cases, 22 were female (8.5/100,000) and 18 were male (7.0/100,000) (Figure 8.1).

Note: The number of cases are indicated in each column.
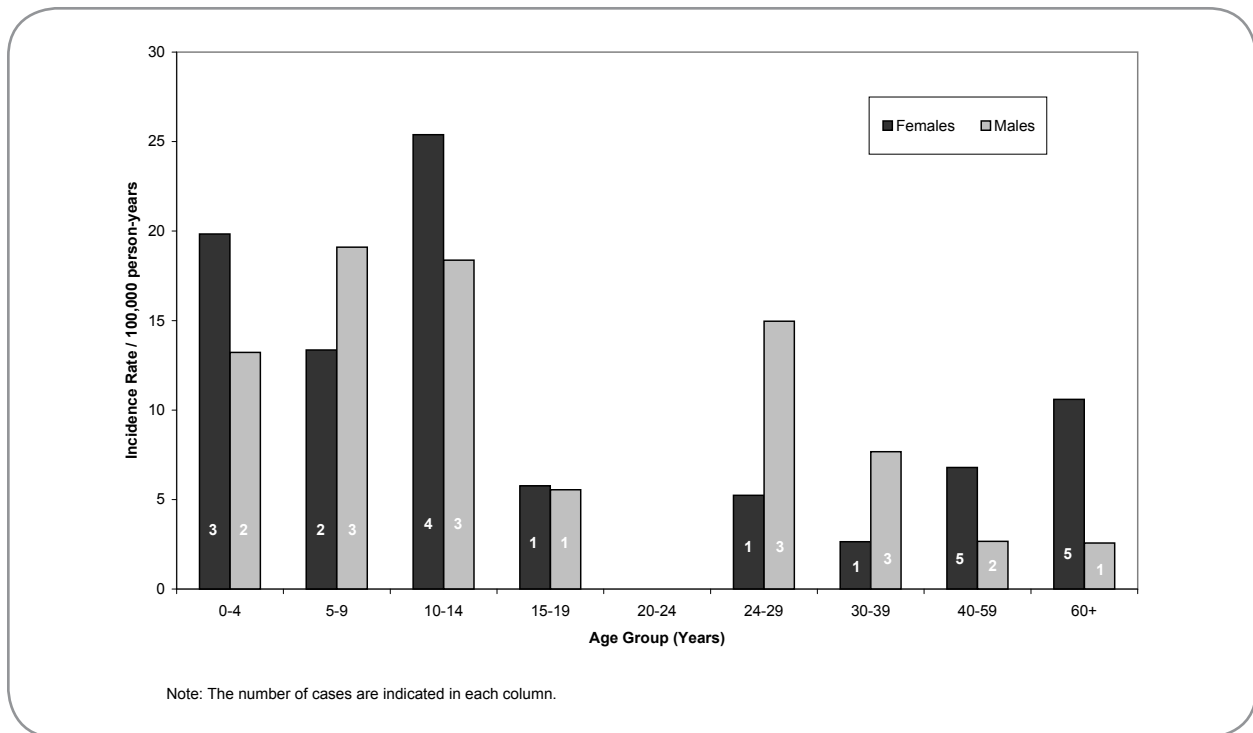
Figure 8.1. Incidence rates of endemic giardiasis cases by gender and age group in Sentinel Site 1 in 2009
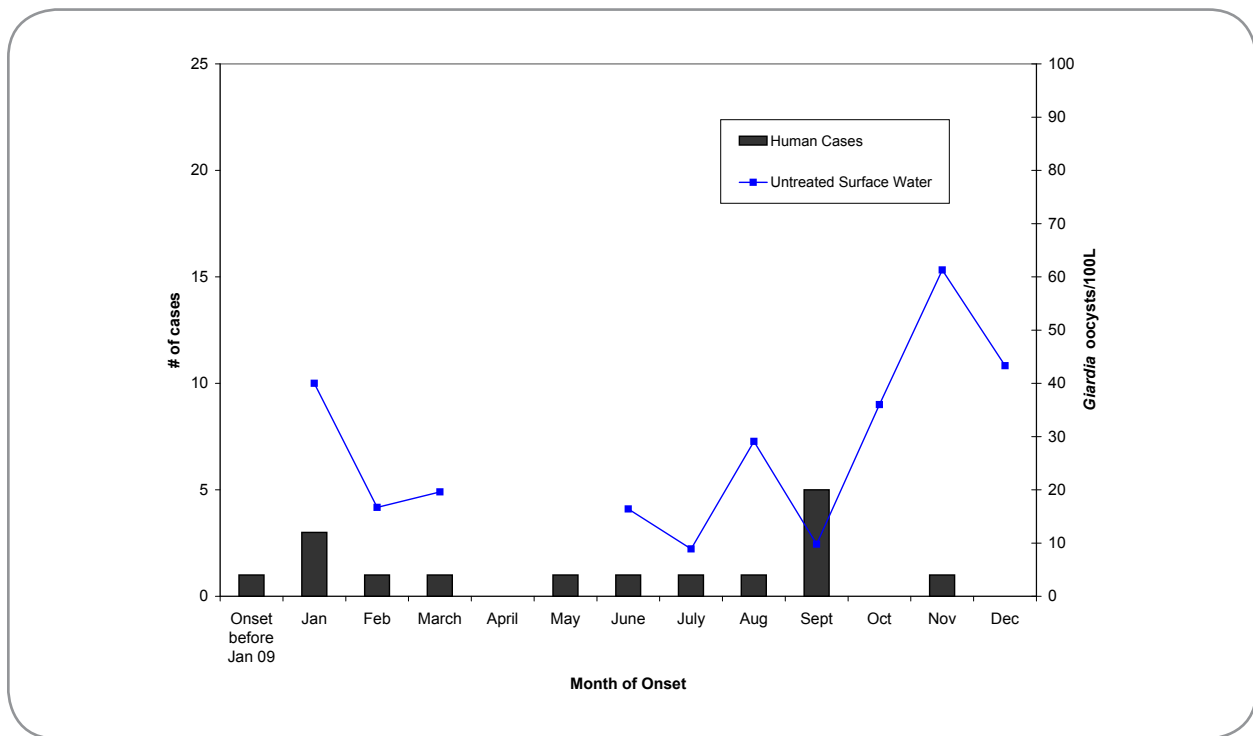
Figure 8.2. Monthly distribution of *Giardia* cases and detection in untreated surface water sampled in Sentinel Site 1 in 2009

The monthly numbers of cases varied from 1 to 5 with the highest reported cases in September, which follows the expected late summer/early fall peak observed with this disease (Figure 8.2) (*10*). Water samples were not collected for April or May of 2009 due to laboratory limitations.

Potential risk factor information for the 25 days prior to the onset of illness was available for 33/40 (83%) of the endemic cases (Appendix B). The *Giardia* cases had higher reported proportions compared to the other enteric cases for the following exposures: drank untreated water, swimming, contact with horses in a farm animal area, and knowing someone with a diarrheal disease the week before illness.

8.1.1 Exposure Surveillance

On-Farm

In 2009, none of the farm manure samples were analysed for *Giardia* (Table 8.1).

Retail Food

In 2009, of the 376 bagged leafy green samples collected, 9 (2%) were positive for *Giardia* by molecular methods. These 9 samples were then tested by microscopy, which detected only one positive sample. This sampling started in March of 2009. Of the 8 samples that were sequenced, 6 were Assemblage B and 2 were Assemblage A, both considered to be infectious to humans. However, these results do not indicate whether the cysts were viable or infective.

Water

Of the 10 samples collected in 2009 just upstream of the drinking water treatment plant intake (site D), *Giardia* was detected in 100% of the untreated surface water samples, (Table 8.1). Further molecular sub-typing was successfully performed on 4 samples, and all were positive for *G. microti* (not considered a human pathogen). The average concentrations of *Giardia* cysts were highest between January and April (Figure 8.2).

TABLE 8.1
***Giardia* detection and sub-typing data for the integrated surveillance activities in Sentinel Site 1 in 2009 (values in brackets refer to data from 2005-2008)**

	Human	Food Animals (Manure)				Retail Food	Untreated Surface Water
	Endemic Cases	Swine	Broiler Chickens	Beef	Dairy	Produce	Grand River
						Bagged leafy greens	5 sample points on Grand River
Microscopic Results		(2005-2006)	(2007-2008)	(2007-2008)	(2005-2006)		(2008)
# tested	(Unknown)	(122)	(126)	(112)	(179)	9[a]	10 (22)
# positive	(48)	(62)	(0)	(72)	(72)	1	10 (D) (21)
% positive		(51%)	(0%)	(64%)	(40%)	11%	100%
PCR Results							
# tested		(122)	(126)	(112)	(179)	376	
# positive		(80)	(12)	(77)	(54)	9	
% positive		(66%)	(10%)	(69%)	(30%)	2%	
Sequencing results							
# samples with sequencing results		(63)	(7)	(73)	(43)	8[b]	4[b]
Assemblage A			(1)		(3)	2	
Assemblage B		(58)	(4)		(18)	6	
Assemblage E		(5)	(2)	(73)	(22)		
G. microti							4 (D)

[a] Culture method
[b] Not all positive samples were sequenced
Water Sampling Locations in Grand River Watershed:
A - Canagagigue Creek
B - Conestogo River
C - Upper Grand River
D - Grand River, near drinking water intake
E - Grand River, near one wastewater treatment plant effluent point

8.2 *Cryptosporidium*

In 2009, there were a total of 20 reported cases of cryptosporidiosis (3.9/100,000 person-years). Of these 20 cases, 3 (15%) were travel-related (0.6/100,000 person-years) and 17 were classified as endemic (3.3/100,000 person-years) (Figure 8.3). In comparison, the annual incidence rates for cryptosporidiosis in 2008 in Canada and Ontario were 2.4/100,000 and 2.6/100,000, respectively (*1*). Of the endemic cases, 4 were female (1.5/100,000) and 13 were male (5.1/100,000).


Note: The number of cases are indicated in each column.
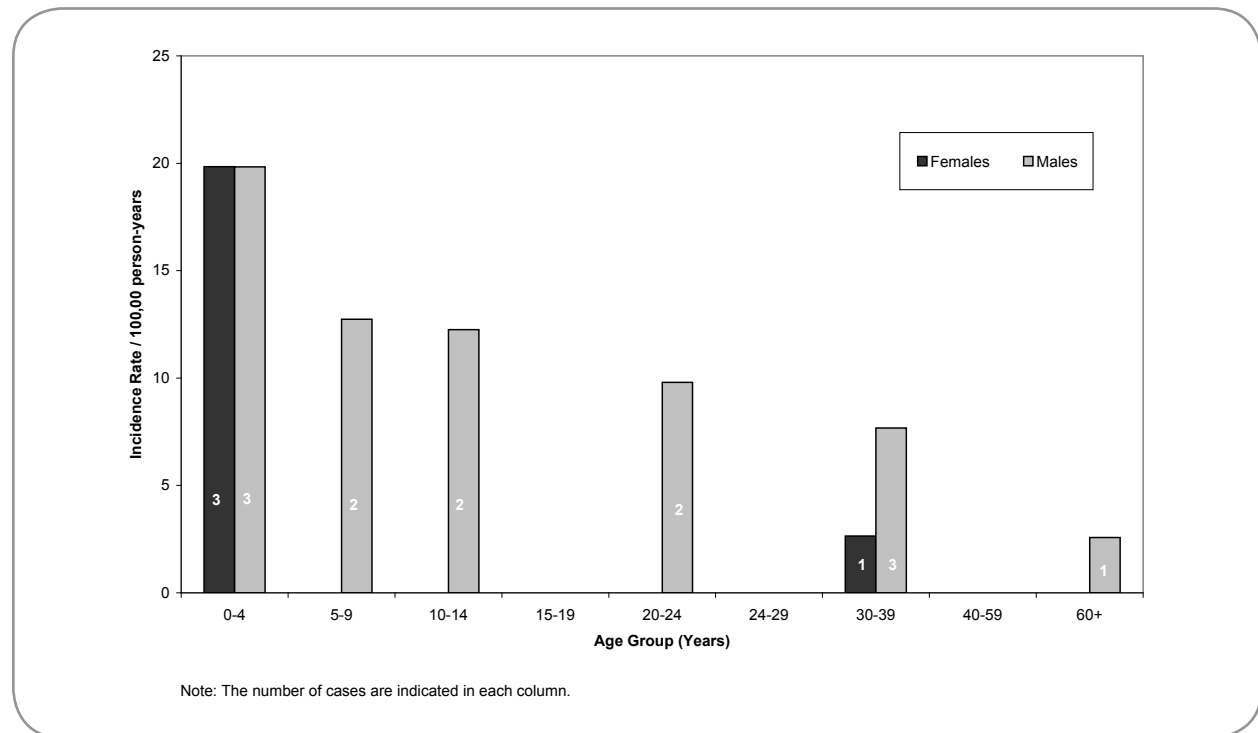
Figure 8.3. Incidence rates of endemic cryptosporidiosis cases by gender and age group in Sentinel Site 1 in 2009

Potential risk factor for the 12 days prior to the onset of illness was available for 13/17 endemic cases (Appendix B). The *Cryptosporidium* cases had higher reported proportions compared to the other enteric cases for the following exposures: using a private well, swimming (in a pool or lake), drinking unpasteurized milk, drinking other unpasteurized products, attending a barbecue, eating in a restaurant, eating meat from private kill, shopping at a butcher shop, living on a farm or in a rural area, on-farm exposure to cats, dogs, cattle, pigs, poultry and sheep, and visiting a farm animal area (horses, cattle, poultry).
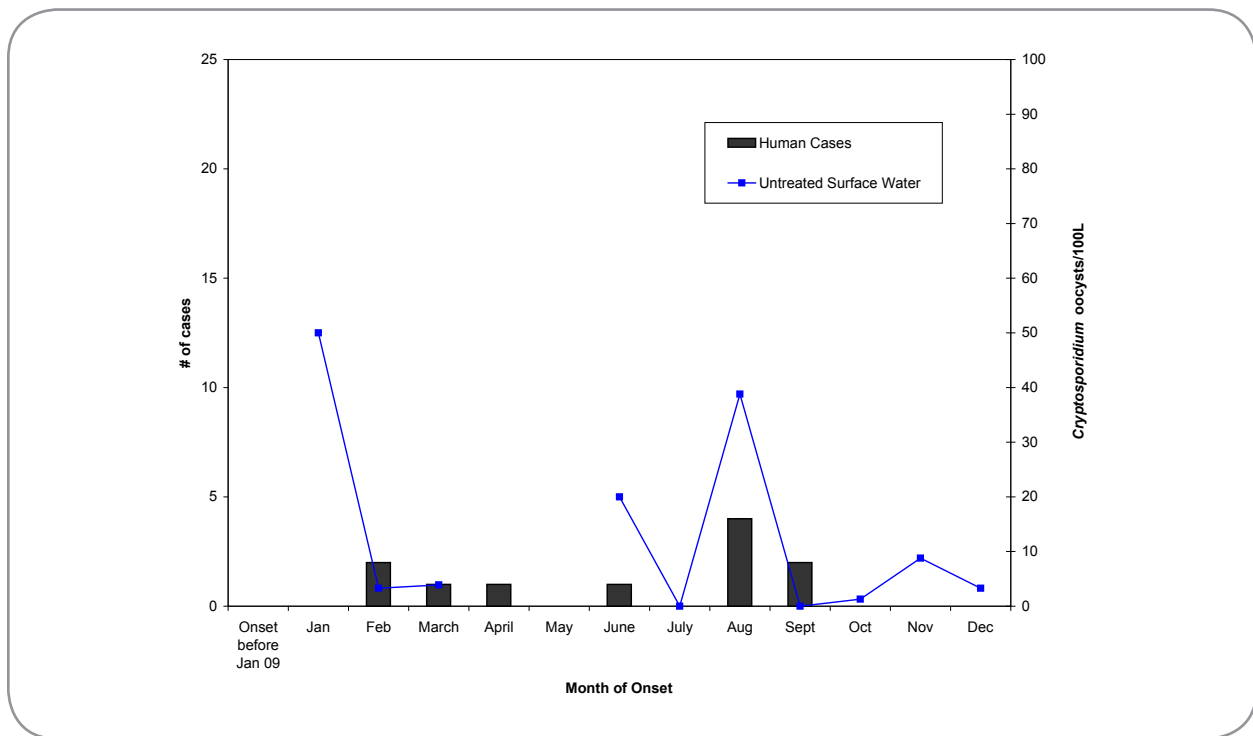
Figure 8.4. Monthly distribution of *Cryptosporidium* cases and detection in untreated surface water sampled in Sentinel Site 1 in 2009

Endemic cryptosporidiosis cases occurred from February to September (Figure 8.4). *Cryptosporidium* oocyst levels remained variable throughout the year. Water samples were not collected for April or May of 2009 due to laboratory limitations.

The average concentration of *Cryptosporidium* oocysts in untreated surface water peaked in March, corresponding with the spring thaw, and fluctuated between 4 and 20 oocysts/100L for the remainder of the year (Figure 8.4). There appeared to be no temporal relationship between the presence of *Cryptosporidium* in untreated surface water and the onset of human cases.

8.2.1 Exposure Surveillance

TABLE 8.2

***Cryptosporidium* detection and sub-typing data for the integrated surveillance activities in Sentinel Site 1 in 2009 (values in brackets refer to data from 2005-2008)**

	Human	Food Animals (Manure)				Retail Food	Untreated Surface Water
	Endemic Cases	Swine	Broiler Chickens	Beef	Dairy	Produce	Grand River
						Bagged leafy greens	5 sample points on Grand River
Microscopic Results		(2005-2006)	(2007-2008)	(2007-2008)	(2006)	2009	(2008)
# tested	Unknown	(122)	(126)	(112)	(179)	32[c]	10 (24)
# positive	15	(54)	(0)	(27)	(14)	23	8 (D) (22)
% positive		(44%)	(0%)	(24%)	(8%)	72%	80%
PCR Results							
# tested		(122)	(126)	(112)	(179)	376	
# positive		(68)	(13)	(31)	(40)	32	
% positive		(56%)	(10%)	(28%)	(22%)	9%	
Sequencing results							
# samples sequenced		(53)	(7)	(28)	(23)	28[d]	7 (D) (12)[e]
C. andersoni[a]			(1)	(27)	(9)		6 (10)
C. baileyi chicken genotype (CB01)							
C. bovis					(2)		
C. cervine[e]							
C. muris[a,b]		(3)	(1)				(1)
C. hominis[a,b]							1 (1)
C. muskrat genotype I (Cluster W 7)							(2)
C. muskrat genotype II (Cluster W 15)							
C. parvum (bovine genotype)[a]		(31)	(6)	(1)	(11)	28	1
C. ryanae[a]					(2)		
C. suis[a]		(1)					
C. cervine genotype							1
C. chipmunk genotype							
C. ferret-like genotype							
C. fox genotype (Cluster W 24)							
C. sp. 2622 host-cattle							
C. skunk genotype							1
C. pig genotype: II[a]		(20)					

[a] Known to be pathogenic to humans
[b] Only found in humans
[c] Culture method
[d] Not all positive samples were sequenced
[e] Some samples have more then one sequencing result, therefore the column total may exceed the total number sequenced

Water Sampling Locations in Grand River Watershed:
A - Canagagigue Creek
B - Conestogo River
D - Grand River, near drinking water intake
E - Grand River, near one wastewater treatment plant effluent point

On-Farm

In 2009, none of the farm manure samples were analysed for *Cryptosporidium* (Table 8.2).

Retail Food

In 2009, molecular methods detected 32 *Cryptosporidium* positive samples (9%) of the 376 bagged leafy green samples collected. Further testing by microscopy found 23 of these 32 samples positive (72%). This sampling started in March of 2009. Of the 28 samples genotyped, *C. parvum* was the only genotype detected.

Water

Fewer samples were analysed in 2009 and restricted to monthly sampling at a point upstream of the drinking water treatment plant intake. Detection of *Cryptosporidium* in 80% of the water samples (all taken at site D) indicates a high prevalence of this potential pathogen in the watershed (Table 8.2). In 2009, *C. andersoni* once again dominated as the most common genotype. It should be noted that *C. andersoni*, while not commonly associated with human infections, has recently been reported in some immunocompetent cases (***11,12***), suggesting that it might indeed be mildly infectious. Both *C. hominis* and *C. parvum*, human-pathogenic strains, were detected in one of the 10 samples tested.

- In the sentinel site, both *Giardia* and *Cryptosporidium* appears to be endemic in untreated surface water. There appears to be no correlation between high levels of *Cryptosporidium* or *Giardia* oocysts in the untreated surface water and human cases (Figures 8.2 and 8.4).

- *C. hominis,* which is host specific to humans, was detected in untreated surface water. *C. andersoni*, although rarely reported in human cases, was also found in untreated surface water.

- This is the first year that bagged leafy greens have been sampled, and results suggest that both human infectious strains of *Giardia* and *Cryptosporidium* are present on this food source. However, these results do not indicate whether the detected protozoa were viable (capable of causing infection).

KEY MESSAGE:

Both *Giardia* and *Cryptosporidium* are continuously present in the Grand River watershed, including both human-infectious and non-human-infectious strains. Further surveillance will determine whether the presence of these two parasites in bagged leafy greens is consistent year to year and a possible route of exposure.

8.3   Cyclosporiasis

Four travel-related (0.77/100,000 person-years) cases were reported in Sentinel Site 1 in 2009.

Cyclosporiasis is not considered to be endemic to Canada. Therefore, active surveillance for *Cyclospora* was not performed for the agriculture and water sources included in the C-EnterNet program. Testing for this parasite was performed on bagged leafy greens. Initial pre-screening by molecular methods identified 2% of the samples positive for *C. cayatenensis*, and by subsequent microscopy confirmation, 56% (5/9) of those samples were positive.

TABLE 8.3
***Cyclospora* detection and sub-typing data for
retail sampling in Sentinel Site 1 in 2009**

Retail Food	
Produce - Bagged Leafy Greens	
Microscopy Results	
# tested	9
# positive	5
% positive	56%
PCR Results	
# tested	376
# positive	9
% positive	2%
Sequencing Results	
# samples with sequencing results	3
C. cayetanensis	3

8.4   Amoebiasis

In 2009, there were a total of 27 reported cases of amoebiasis (5.2/100,000 person-years). Of these 27 cases, 14 were travel-related (2.7/100,000 person-years) and 13 were classified as endemic (2.5/100,000 person-years). Of the endemic cases, 8 were female (3.1/100,000) and 5 were male (1.9/100,000) (Figure 8.5).

Amoebiasis was removed from national surveillance as of January 2000 (*13*); therefore, comparative incidence data cannot be provided for Canada.

Note: The number of cases are indicated in each column.
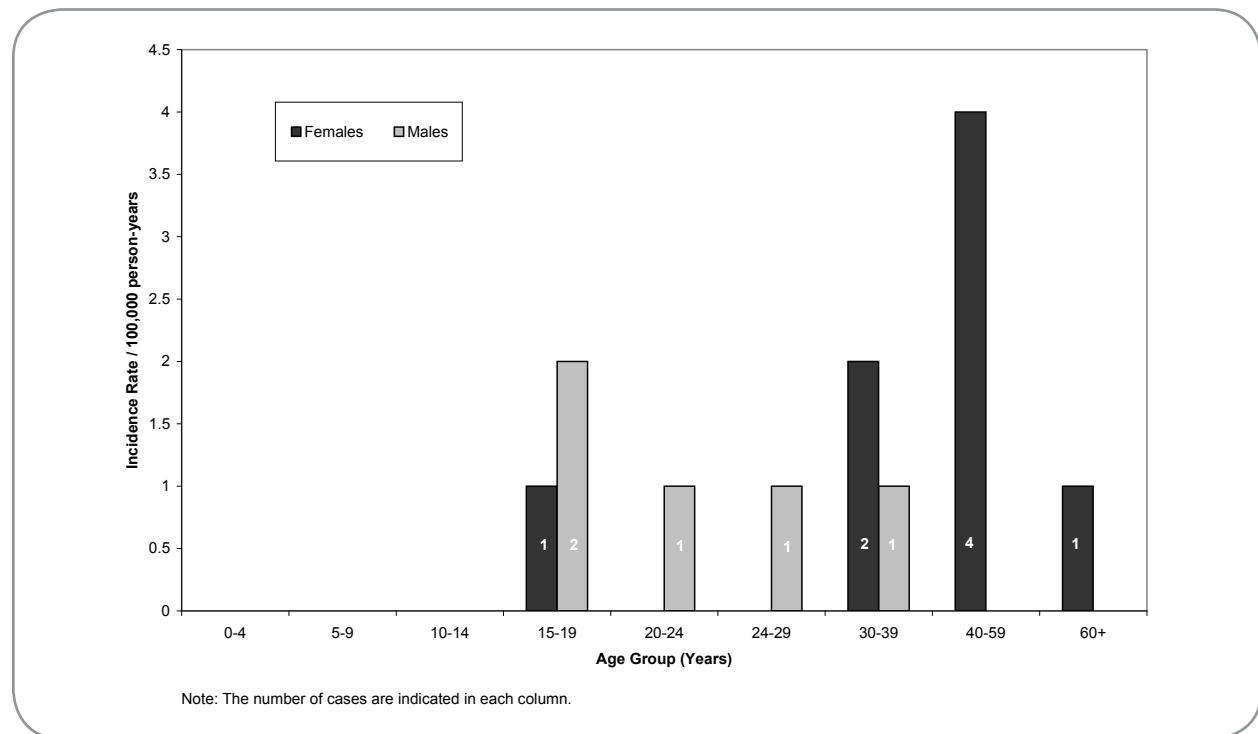
Figure 8.5. Incidence rates of endemic amoebiasis cases by gender and age group in Sentinel Site 1 in 2009

Potential exposure information for the 7 days prior to the onset of illness was available for 12 of the 13 cases (92%) (Appendix B). The following proportions were higher for the amoebiasis cases compared to other enteric cases: drank untreated water and shopping at a butcher shop.

Entamoeba is a human intestinal pathogen. While not considered a zoonotic agent, *Entamoeba* has been known to infect dogs (*14*). It was not assessed in the various exposure sources (food, agriculture and water) in Sentinel Site 1.

9. *Shigella*

9.1   Human Cases

In 2009, there were a total of 8 reported cases of shigellosis (1.5/100,000 person-years). Of these, 1 was travel-related (0.2/100,000 person-years) and 7 were classified as endemic (1.4/100,000 person-years). Of the endemic cases, 4 were female and 3 were male. In comparison, the annual incidence rates for shigellosis in 2008 in Canada and Ontario were 2.26/100,000 and 1.86/100,000, respectively (**1**).

9.2   Exposure Surveillance

For the retail component, testing was initiated in March 2009 on produce (ie: bagged leafy greens). One sample was positive for *Shigella* by screening molecular PCR, but did not yield a positive by culture, therefore viability could not be determined.

TABLE 9.1
***Shigella* detection in human cases and on retail food**
(bagged leafy greens) in Sentinel Site 1 in 2009

	Human		Retail Food
	Endemic	Outbreak	Produce
Detection			Bagged Leafy Greens
Culture Results			
# samples tested	Unknown	Unknown	1
# positive	7	0	0[a]
% positive			0%
Molecular Results			
# samples tested			376
# positive			1
% positive			0.3%

[a] Culture only performed on the single PCR positive

10. Viruses

10.1 Human Cases

Human infections of Norovirus or Rotavirus are not routinely laboratory-confirmed and are not reportable in Sentinel Site 1, except for outbreaks. Norovirus outbreaks became nationally notifiable in 2009 with varying stages of implementation among the reporting provinces and territories.

10.2 Exposure Surveillance

Testing for Norovirus and Rotavirus was initiated on bagged leafy greens in March, 2009. Norovirus was detected by molecular PCR in 5% of the samples (Table 10.1), while Rotavirus was detected by molecular PCR in less than 1% of the samples (Table 10.2). Of the 19 norovirus strains confirmed, 15 belonged to genogroup I (GI) and 6 to genogroup II (GII). All were strain types known to be human pathogens. The group A rotavirus was not subtyped; group A rotaviruses can be human or animal pathogens. This indicates, but does not prove, that there may be infectious enteric viruses on packaged, ready-to-eat leafy greens that could cause gastroenteritis in the Canadian population.

TABLE 10.1
Norovirus detection on bagged leafy greens in Sentinel Site 1 in 2009

Bagged leafy greens	Norovirus
PCR Results	
# tested	376
# positive	19
% positive	5%
Genotype results[a]	
# samples with sequencing results	19
GI	15
GII	6

[a] Two samples had more than one genotype detected

TABLE 10.2
Rotavirus detection on bagged leafy greens in Sentinel Site 1 in 2009

Bagged leafy greens	Rotavirus
PCR Results	
# tested	376
# positive	1
% positive	0.30%
Genotype results	
# samples with sequencing results	1
Group A	1

11. Temporal Variations

Identifying temporal trends or seasonal and other cyclical variations over time is a key function of health surveillance. It allows for the interpretation of the current state of health issues in the context of the historical background and to forecast future problems and related consequences.

11.1 Temporal Variations in Enteric Disease Incidence

Seasonal variation

The monthly counts of sporadic, non travel-related cases since C-EnterNet's implementation in Sentinel Site 1 from June 2005 to December 2009 (Figure 11.1) visually show seasonal patterns of disease occurrence, with more cases during summer or fall for all diseases, with the exception of yersiniosis and amoebiasis.

A negative binomial regression model was used for each disease separately to formally test for both annual and seasonal variations. Full years of data from 2006-2009 were used. The seasonal variation was based on month for the two most frequent diseases (campylobacteriosis and non-typhoidal salmonellosis) and by quarter (winter: December to February; spring: March to May; summer: June to August; fall: September to November) for the other diseases (giardiasis, amoebiasis, cryptosporidiosis, yersiniosis, and verotoxigenic *E. coli* (VTEC) infections).

With regards to seasonal variations, the following results were statistically significant ($p < 0.05$):

- Campylobacteriosis was higher in June, July, August, and September compared to any of the other months;

- Non-typhoidal salmonellosis was higher in May, July, August and September compared to any other months;

- Cryptosporidiosis was higher in summer compared to winter;

- VTEC infections were higher in summer compared to spring and to fall (there were no cases in winter).

Annual variation

To compare the annual variations in endemic cases of enteric diseases, the annual incidence rates were computed for each disease and plotted relative to the annual incidence rate observed for the first full year of C-EnterNet's implementation in Sentinel Site 1, i.e. year 2006 (Figure 11.2). The only monotonous (either steady increase or steady decrease) trends were a noted decrease in yersiniosis and VTEC infections. In addition, the non-typhoidal salmonellosis incidence rate was lower in 2006 compared to any other years. The annual incidence rates for the other diseases have fluctuated above and below the 2006 incidence rate without specific patterns.
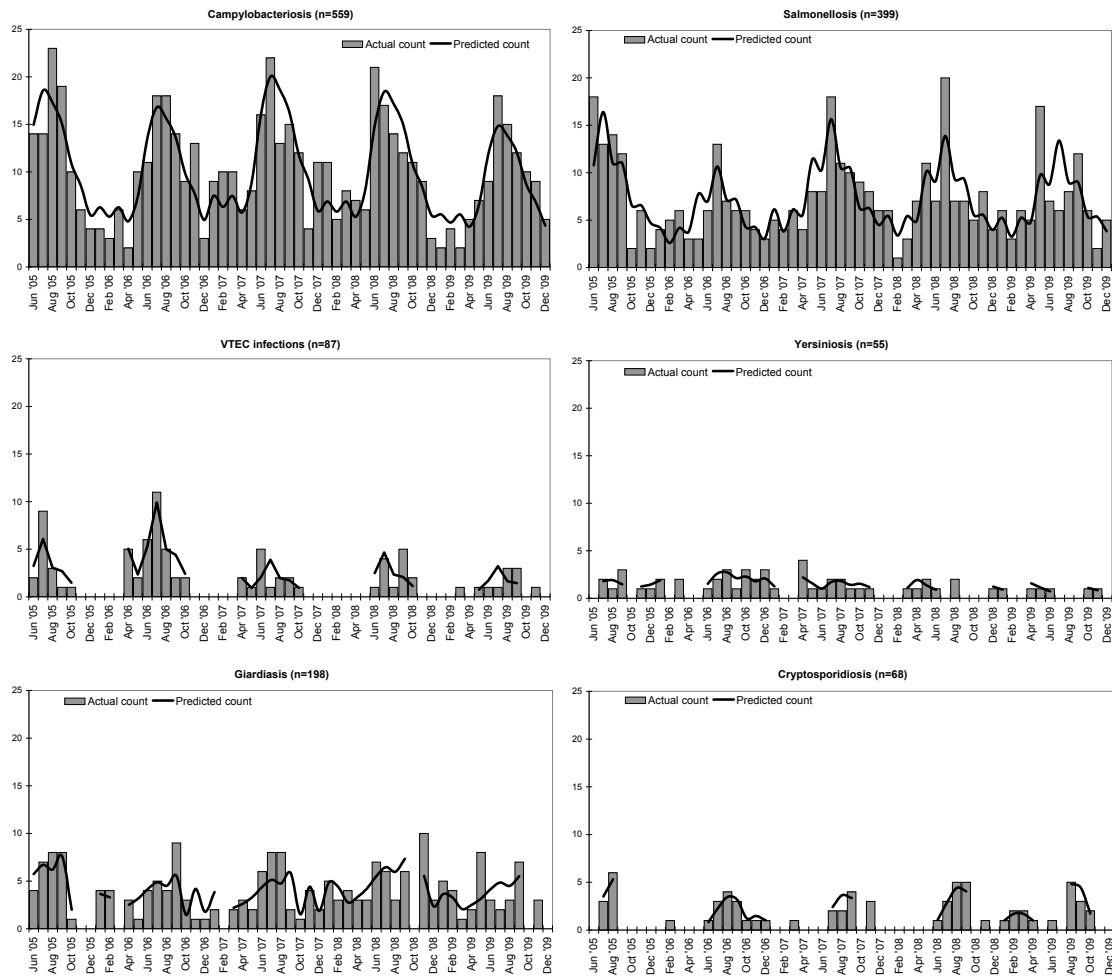
Figure 11.1. Monthly counts (based on onset dates) of sporadic, non travel-related cases reported in Sentinel Site 1 from June 2005 to December 2009 for selected enteric diseases and the smoothed predicted counts according to a fitted negative binomial regression model
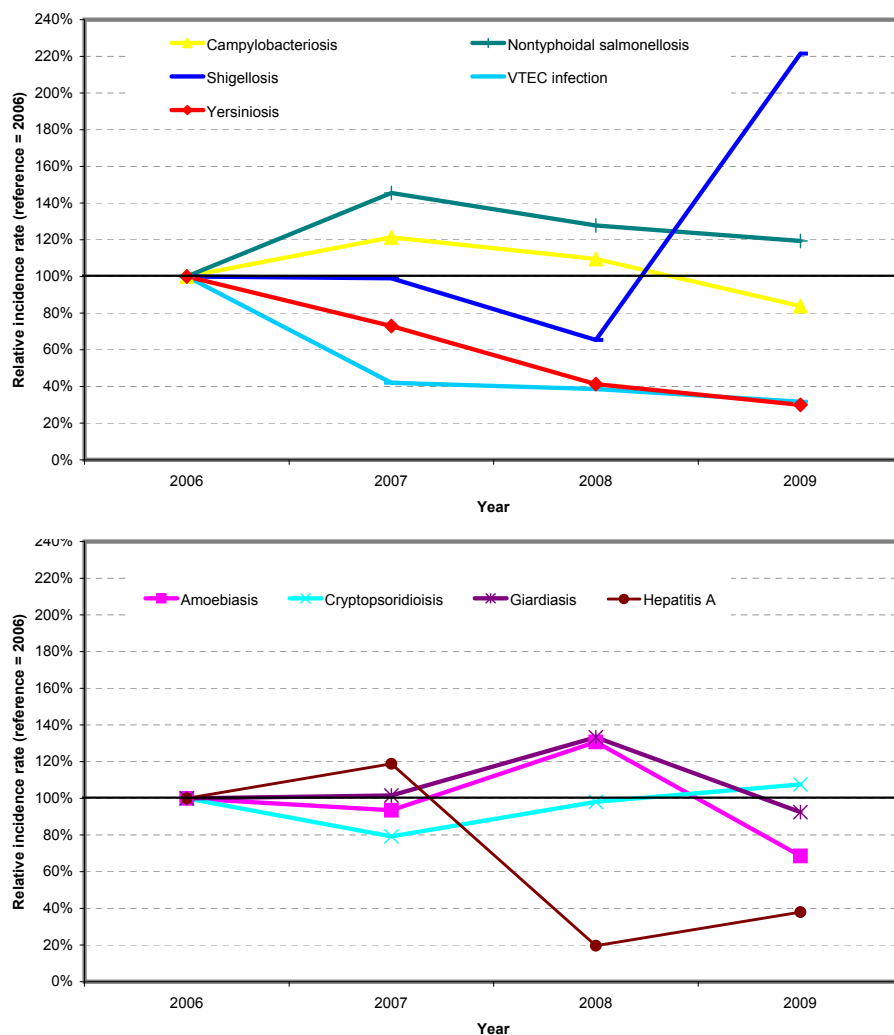
Figure 11.2. Changes in annual incidence rates of sporadic, non travel-related cases of reportable enteric diseases over the years relative to the first full year of enhanced surveillance implementation in Sentinel Site 1 (2006)

The statistically significant results (p<0.05) of the negative binomial regression model accounting for season (month or quarter) (see above) with regards to years are:

- Campylobacteriosis was greater in 2007 compared to any other years;

- Yersiniosis was lower in 2009 compared to 2006;

- VTEC infections were lower in 2007 compared to 2006 and lower in 2009 compared to 2006.

The 2009 annual incidence rates for those diseases were compared to their respective incidence rate observed in the first full year of C-EnterNet's implementation in Sentinel Site 1 (i.e., 2006) with the 95% confidence interval around these incidence rate ratios (Figure 11.3). It shows that VTEC infection and yersiniosis incidence rates were statistically smaller in 2009 compared to 2006, with at decrease by 68% and 70% of their 2006 value, respectively. No other statistically significant differences between 2009 and 2006 were observed.
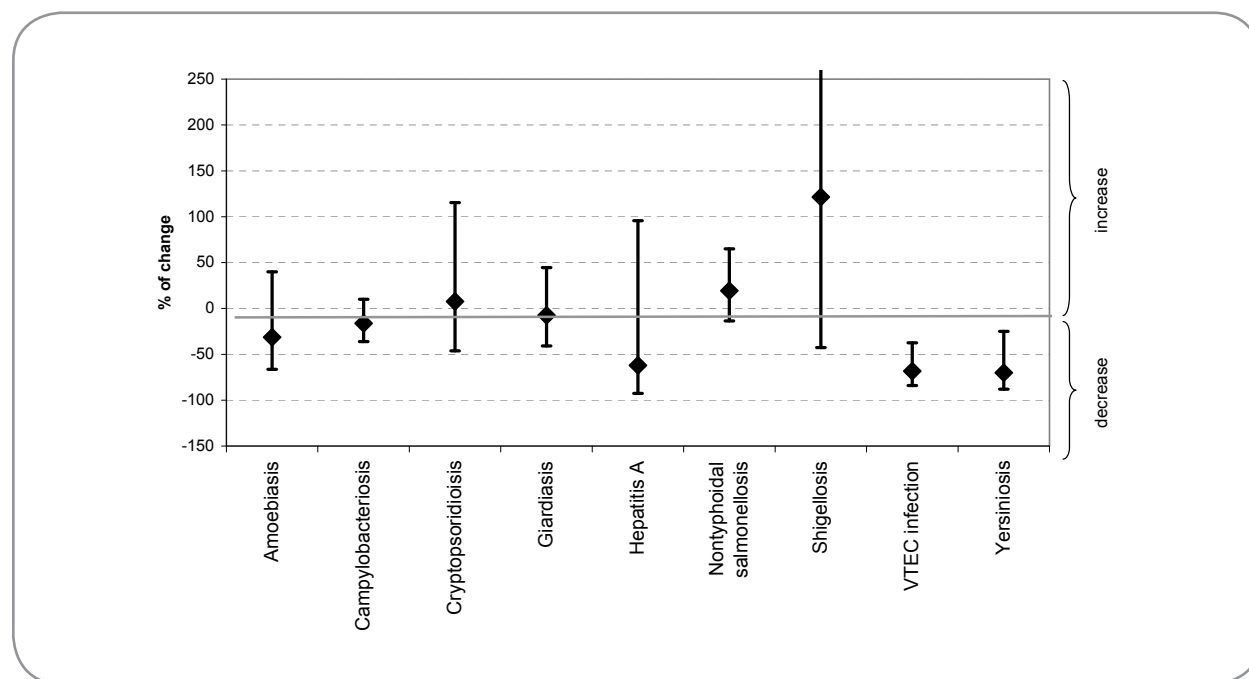
Figure 11.3. Changes (with 95% confidence interval) in annual incidence rates between the year 2009 and the first full year of the enhanced surveillance implementation in Sentinel Site 1 (2006) for sporadic, non travel-related cases of reportable enteric diseases

To minimize the influence of any year used as reference in comparing years for variations, the 2009 annual incidence rates were also compared to their respective average incidence rate observed over the first 3 full years of C-EnterNet's implementation in Sentinel Site 1 (i.e., 2006-2008, inclusive) with the 95% confidence interval around these incidence rate ratios (Figure 11.4). It shows a statistically significant decline for campylobacteriosis and yersiniosis, whereas the decline for VTEC infection was closed to significance. Shigellosis in 2009 was relatively high, but its incidence rates have a large confidence interval due to the few number of cases reported (7 in 2006 vs. 2 in 2007 vs 3 in 2008).
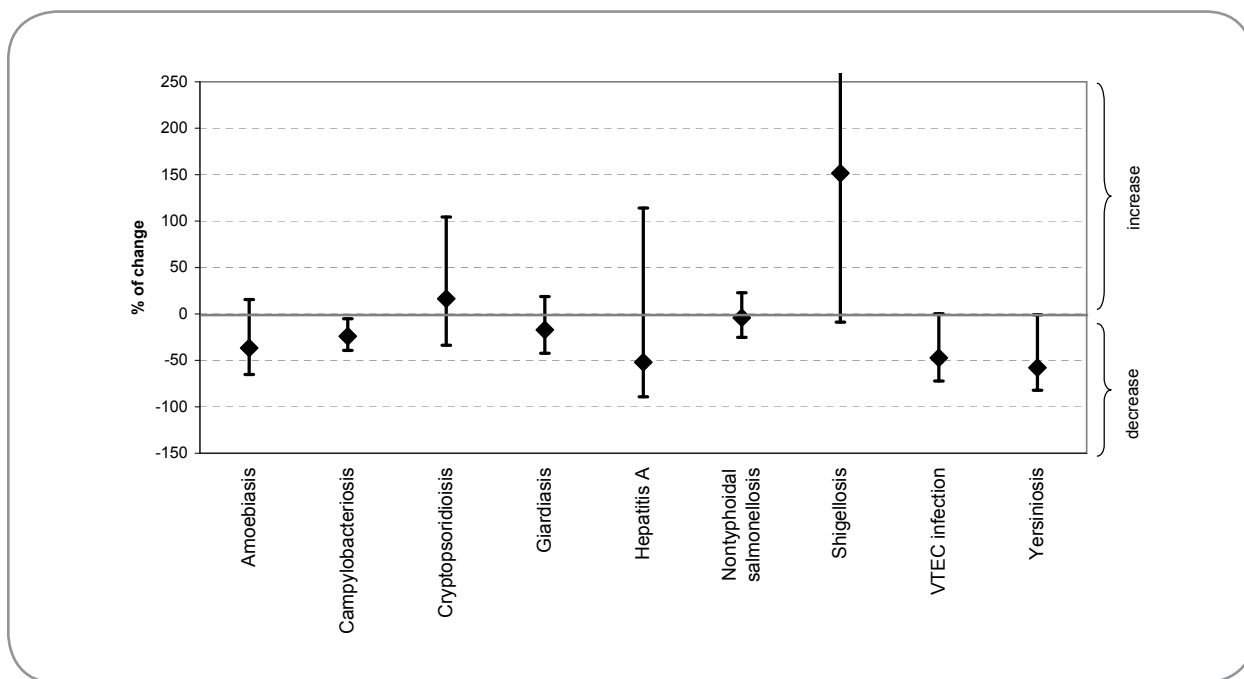
Figure 11.4. Changes (with 95% confidence interval) in annual incidence rates between the year 2009 and the first 3 full years of the enhanced surveillance implementation in Sentinel Site 1 (2006-2008, inclusive) for sporadic, non travel-related cases of reportable enteric diseases

11.2 Temporal Variations in Exposure Source

Agriculture Component

The detection of enteric pathogens on farms represents an environmental exposure source. Each month 2 to 3 farms per commodity are enrolled and visited for a total of approximately 30 farms per commodity per year. The visit involves the administration of a short management survey and sampling of 3 fresh pooled manure samples from different age groups of animals and one stored manure sample.

Results are presented at the sample level (Figure 11.5). The prevalence of *Campylobacter* increased significantly ($p<0.05$) in swine, dairy and beef farms in 2008 and 2009 compared to 2007 and 2006 and is most likely due to the implementation of a more sensitive laboratory methodology at the beginning of 2008, rather than a true prevalence increase. In 2009, the prevalence of *Salmonella* increased significantly on dairy farms and decreased significantly on broiler chicken farms at the sample level, but not at the farm level.
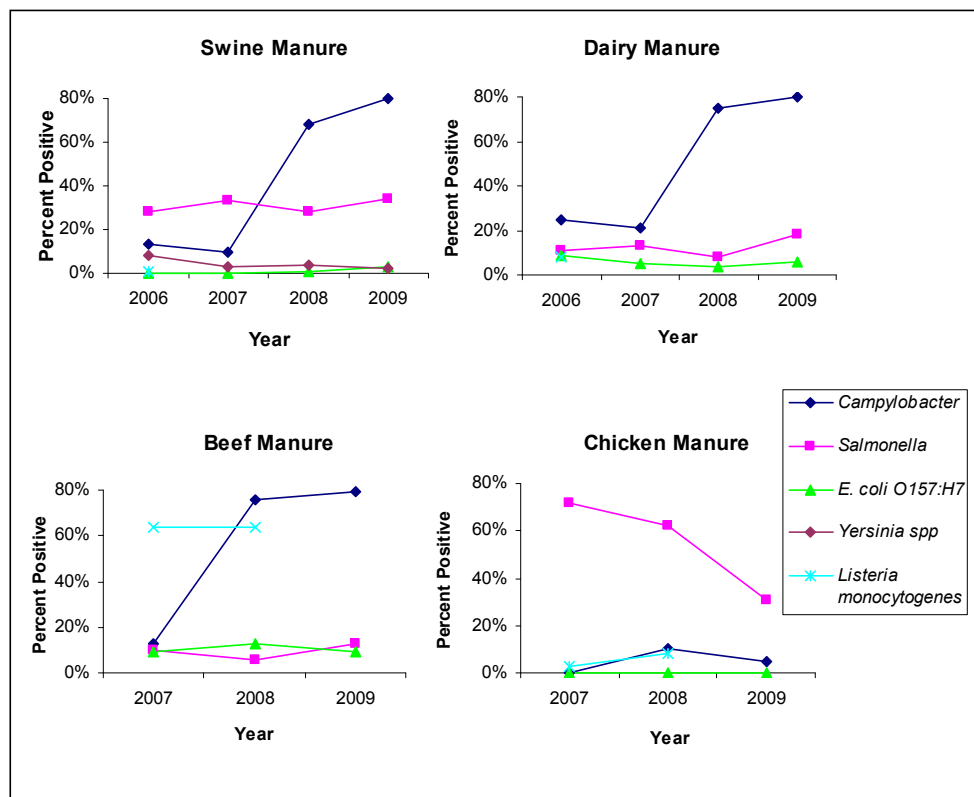
Figure 11.5. Annual variations in pathogens detected from manure samples in Sentinel Site 1, 2006-2009

Water Component

Since 2005, five sites along the Grand River have been sampled for exposure surveillance within the C-EnterNet sentinel site to understand the dynamics of enteric pathogens in the environment and their transmission from both point and non-point sources within the watershed.

Potential annual and seasonal changes are shown in Figures 11.6 and 11.7, respectively. Such potential effects on the probability of a sample to be positive were tested using a conditional logistic regression model (with a general estimation equation) for various pathogens between winter 2006 and fall 2009. The repetition of the sampling at the same five sites along the river was considered in the model. No seasonal effect was observed in the data. Some statistically significant year effects ($p<0.05$) were observed for *Campylobacter*, *Salmonella*, and *Yersinia:*

- Prevalence of *Campylobacter* in all five sites was higher in 2009 compared to 2006;

- Prevalence of *Salmonella* in all five sites was higher in 2009 compared to 2007;

- Prevalence of *Yersinia* was higher in all five sites in 2009 compared to 2006 though this is attributed to method changes (Appendix C).

The statistical model could not be run for *E. coli* O157:H7 because of the low number of positive samples or for *Giardia* and *Cryptosporidium* because of the low number of negative samples.
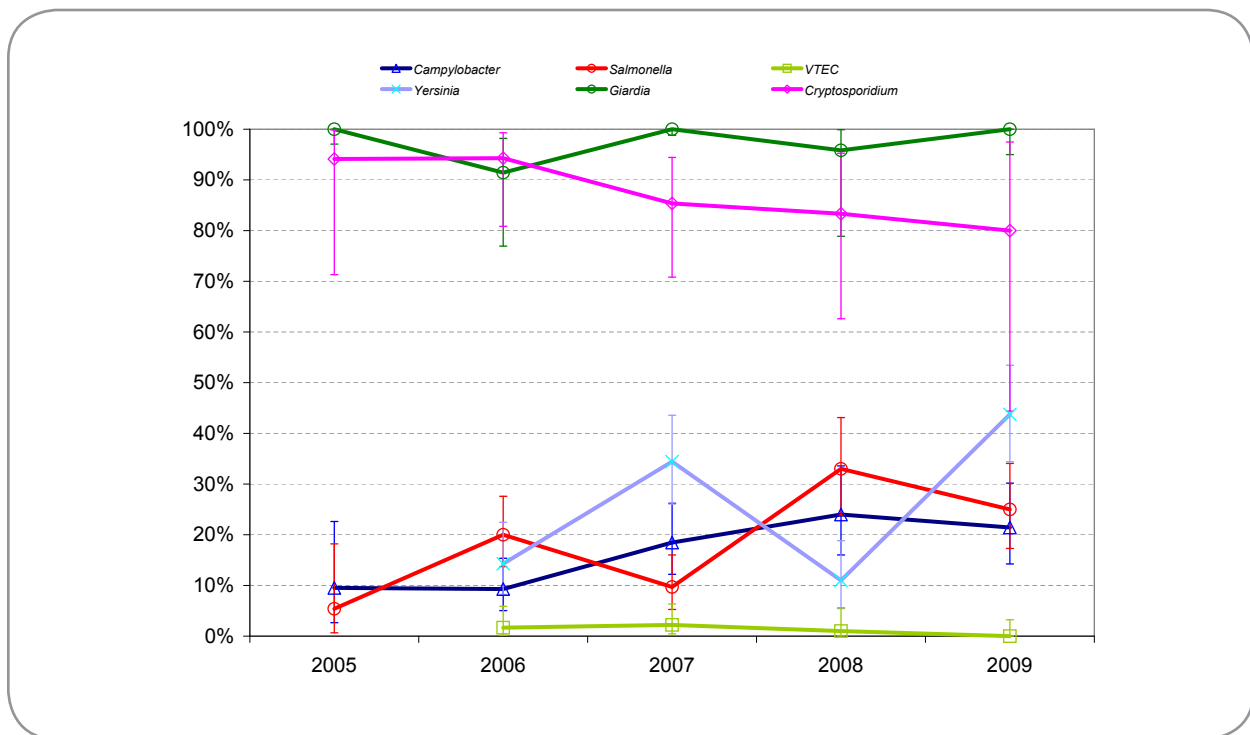
Figure 11.6. Proportion by year (with 95% confidence interval) of positive untreated surface water samples tested by culture method for selected enteric pathogens in Sentinel Site 1 between June 2005 and December 2009
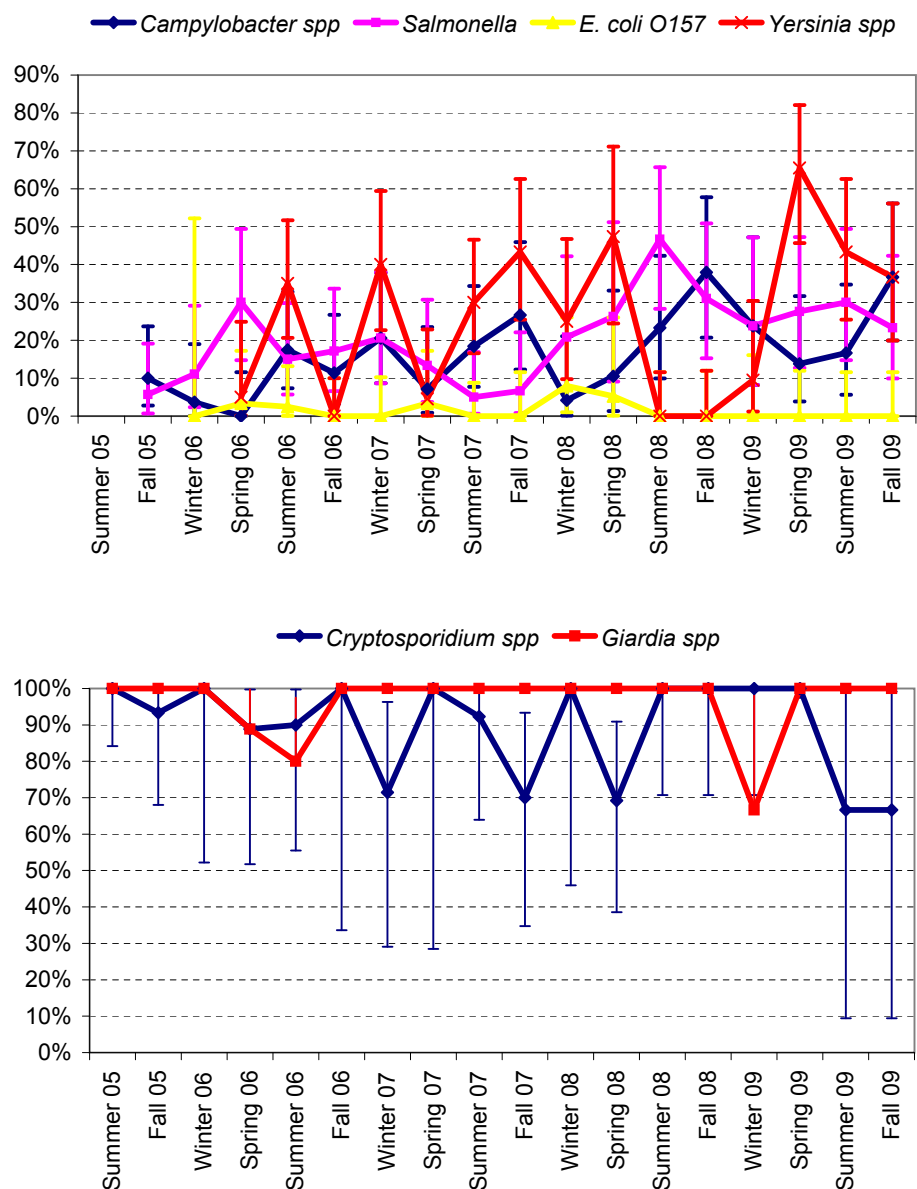
Figure 11.7. Proportion by quarter (with 95% confidence interval) of positive untreated surface water samples tested for selected enteric pathogens by culture method in Sentinel Site 1 between Summer 2005 and Fall 2009 (winter: December to February; spring: March to May; summer: June to August; fall: September to November)

Retail component

Since mid-2005, C-EnterNet has systematically sampled fresh raw pork, chicken and beef from randomly selected grocery stores within the sentinel site on a weekly basis.

Figure 11.8 and Figure 11.9 show the yearly and quarterly distribution of positive raw retail samples (with 95% confidence intervals) from June 2005 to December 2009. Differences between years and between quarters. Quarters were defined as: winter: December to February; spring: March to May; summer: June to August; fall: September to November. They were tested using a conditional logistic regression model (with a general estimation equation) for each pathogen and for each kind of meat separately between winter 2006 and fall 2009. To respect the sampling scheme of the active monitoring put in place for food at retail, the type of store (large vs. small) was included in the model as a covariate and re-sampling within the same store was considered a repetition and was set as such in the statistical algorithm. The following results are significant at p<0.05:

- Chicken meat was more often contaminated by *Campylobacter* spp in 2009 compared to 2007 and to 2006;

- Chicken meat was contaminated by *Campylobacter* spp the most often in fall compared to all other quarters and the less often in winter compared to all other quarters;

- Chicken meat was less often contaminated by *Listeria monocytogenes* in 2009 compared to 2007;

- Pork meat was more often contaminated by *Yersinia* spp in 2009 compared to all other years; in addition it was less contaminated in 2007 compared to 2006;

- Pork meat was more often contaminated by *Yersinia* spp in summer compared to all other quarters;

- Ground beef meat was less often contaminated by *Listeria monocytogenes* in 2009 compared to 2008.

The type of store was only observed to be significant for the prevalence of *Listeria monocytogenes* on pork meat (p<0.001). We were 7.4 times more likely to find a positive *Listeria* result on meat samples purchased from a small store versus a large store (95%CI: 2.7 – 20.2). With a more liberal p-value, the size of store had the same effect on the prevalence of *Listeria monocytogenes* on chicken meat (p=0.07).

Figure 11.8. Proportion by year (with 95% confidence interval) of retail meat positive for selected enteric pathogens in Sentinel Site 1, June 2005 to December 2009

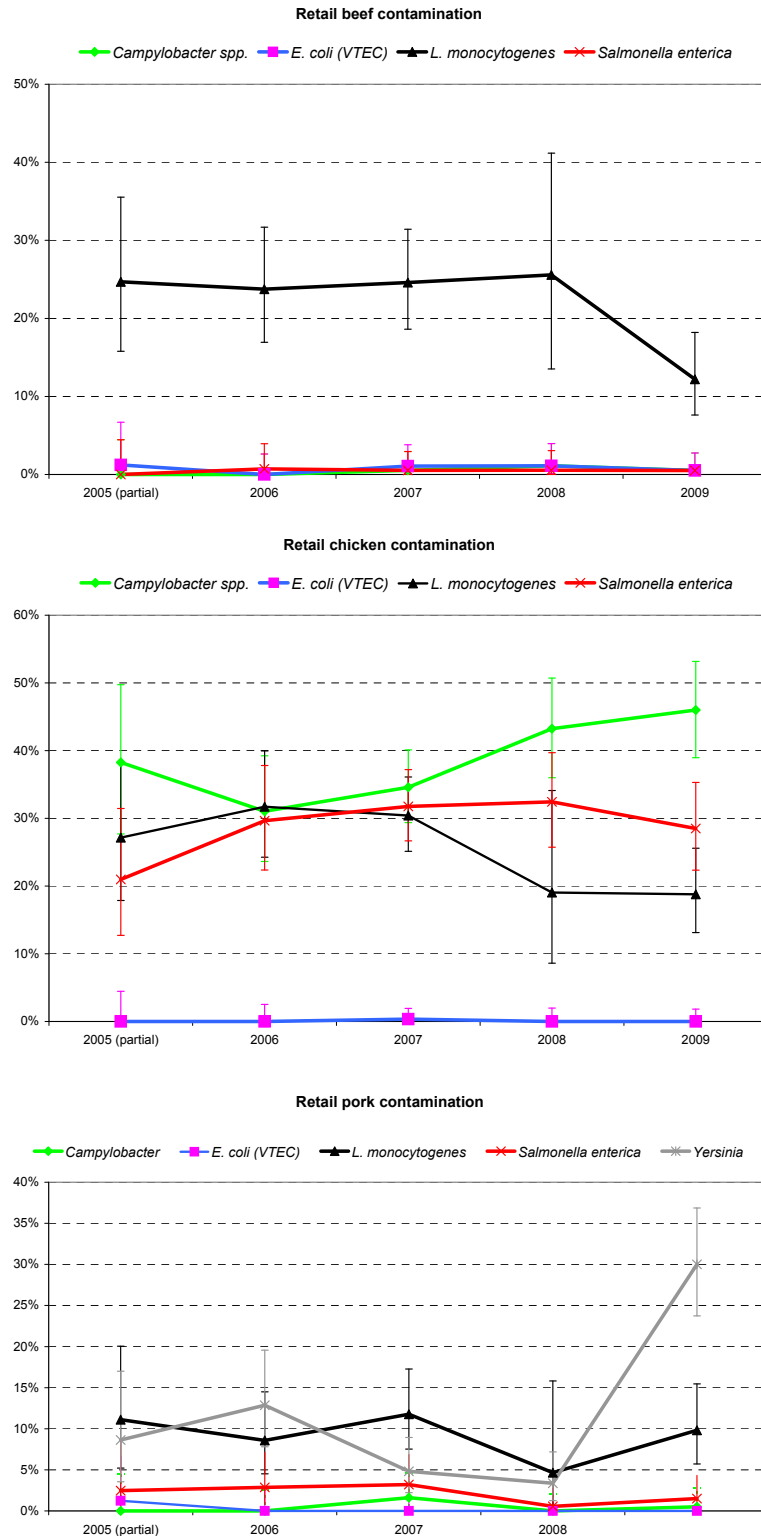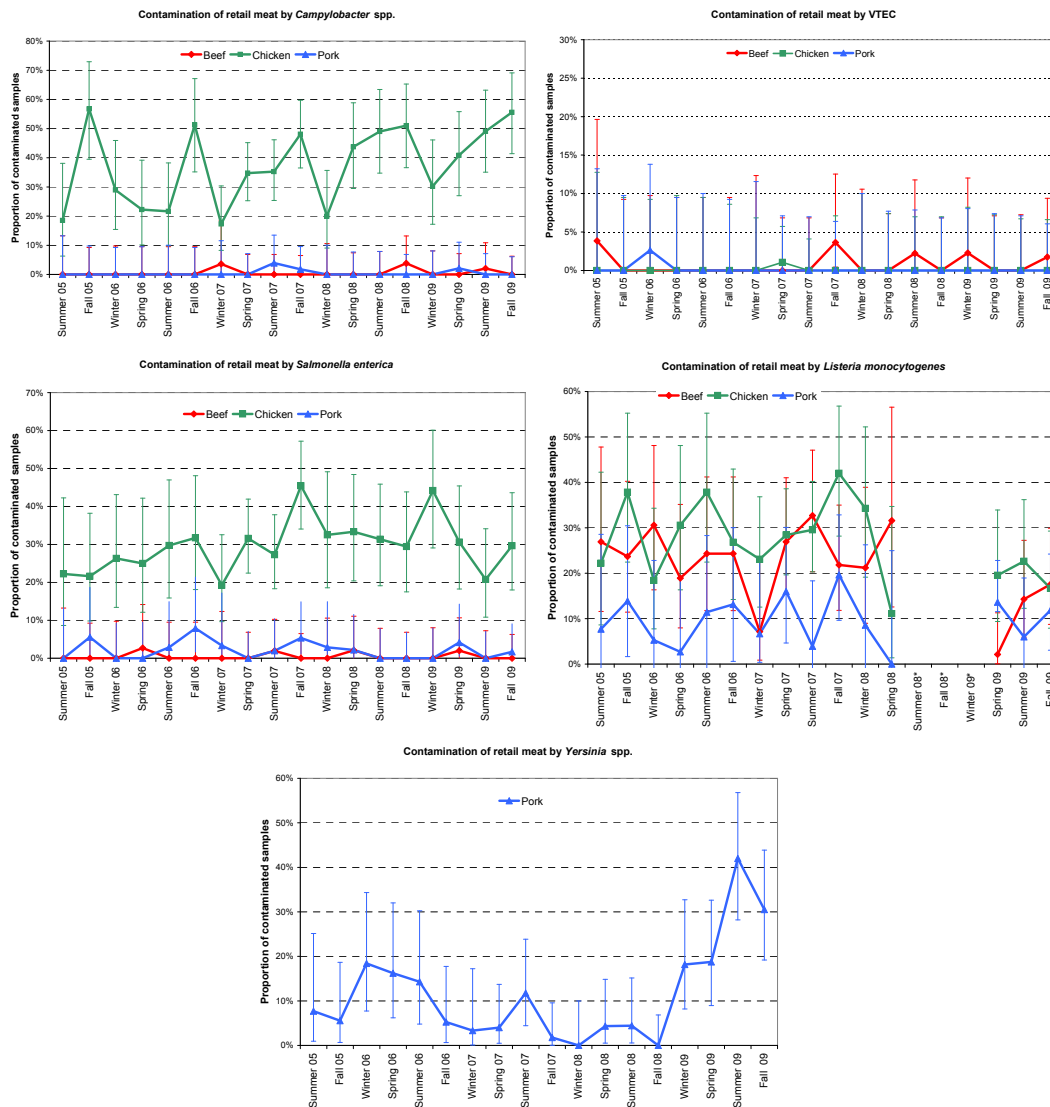**Figure 11.9.** Proportion by quarter (with 95% confidence interval) of retail meats positive for selected enteric pathogens in Sentinel Site 1 between June 2005 and November 2008 (winter: December to February; spring: March to May; summer: June to August; fall: September to November).

* Testing for *Listeria* was not conducted during Summer 2008, Fall 2008 and Winter 2009.

11.3 Association between temporal variations in human incidence and in contamination of exposure sources

Whether, and to what extent, a change over time in the contamination of an exposure source by a given pathogen is linked to a change over time in the incidence of the disease caused by this pathogen in the human population is a fundamental question in the context of an integrated surveillance system. The quantification of such an association has two direct applications, with regards to the prevention and control of human enteric diseases. First, from a forecasting perspective, once the association has been quantified over a given period of time, the change over time in the exposure source (increase or decrease) may predict the corresponding change in human incidence, which in turn will inform the decision about the necessity of new or improved prevention or control measures. Second, in the assessment of the impact of prevention or control measures that are implemented, the observation of a decrease over time of the contamination of the targeted exposure source by the pathogen and a corresponding decrease in the human incidence of that disease will provide some evidence for the effectiveness of the intervention.

Because of the continuous and integrated surveillance design at the sentinel site level, C-EnterNet provides the capacity to quantify these associations between the temporal trends in the human incidence and the exposure sources. As an example, Figure 11.10 displays the evolution in the burden (disease) and prevalence (exposure contamination) of *Campylobacter* spp and *Salmonella enterica* in the Sentinel Site 1. For *Campylobacter*, this figure illustrates an increasing trend in the contamination of retail chicken (although this is attributed to the sampling change, from skin-on to skin-off chicken breasts, see Appendix C) and a greater increase in the surface water contamination, whereas the incidence of human campylobacteriosis has been relatively stable, with a declining trend from 2007. For *Salmonella*, the contamination of retail chicken has been very stable; though levels on retail pork and in untreated surface water varied. Conversely, an increase is observed in the human incidence in 2007 and then showed a slow decline.

The results and interpretations from Figure 11.10 are preliminary. A more in-depth analysis of the association between the incidence of human disease and the exposure source contamination must be more focussed (i.e., considering a specific pathogen species or subtype (serotype, etc)) and must consider source attribution (i.e., the proportion of cases that are attributable to each specific exposure source). Such analysis has been undertaken just recently (*15*). The systematic and continuous collection of data from human cases and from all or, at least, the most important sources is a prerequisite to analyze the temporal variations in human incidence in relation to the ones in the relevant sources. The full C-EnterNet surveillance system with 5 or 6 sentinel sites across Canada was designed for that purpose. Its expansion beyond the two current sentinel sites will help to fulfill the data requirements necessary to quantify the associations between cases of enteric disease and likely sources in Canada.
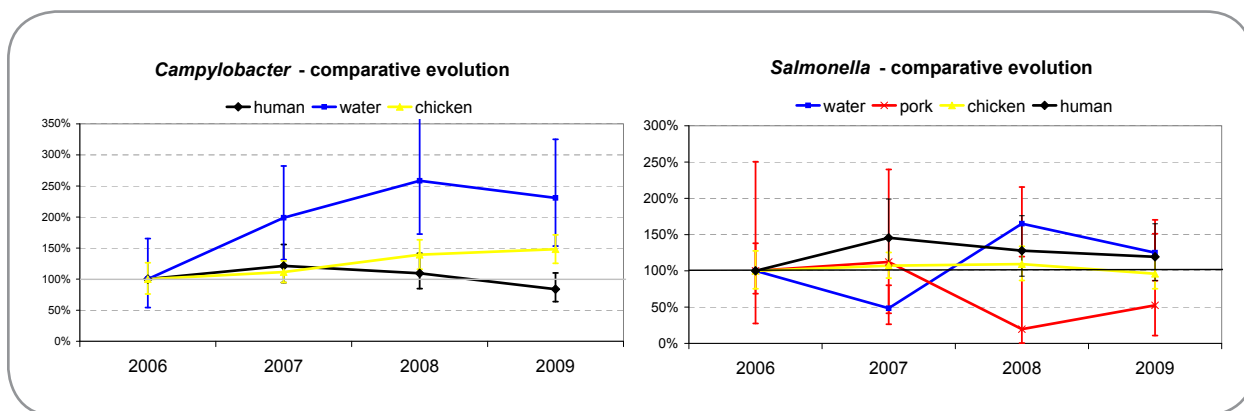
Figure 11.10. Comparative evolution of the temporal changes over time in the human incidence and some selected exposure source contamination for *Campylobacter* and for *Salmonella*. For each exposure source, the contamination and its 95% confidence interval is relative to the value observed for the year 2006. Similarly, the human line shows the incidence rate ratio and it 95% confidence interval with the incidence rate for 2006 as the reference.

12. Source Attribution

The C-EnterNet surveillance program has two core objectives:

- Surveillance: detect changes in trends of human enteric disease incidence and pathogen exposure levels from food, animal and water sources;

- Human illness source attribution: determine the proportion of human cases that are due to water, food and animal contact (*16*).

While still in the developing stages of the surveillance program, and one sentinel site, the source attribution activities are limited in their scope and impact. However, the program is still planning and implementing several projects to refine methodologies and develop preliminary estimates with respect to source attribution to inform food and water safety policy, and the prevention and control of human infectious gastrointestinal illness in Canada (Table 12.1).

Source attribution activities are being pursued in some countries across the world. Based on previous and current research in this area by international food safety experts, several broad methodological approaches are advocated to generate estimates of human illness attribution, including:

- Microbial subtyping approach

- Quantitative microbial risk assessment (QMRA)

- Comparative exposure assessment

- Analysis of data from outbreak

- Case-control studies

- Intervention studies

- Expert elicitation

Each method has its specific advantages and limitations, and experts on source attribution have concluded that none of the currently available methods yields accurate estimates for source attribution on its own. The approaches are quite different and thus address slightly different questions. While still in the expansion phase of the surveillance program, however, the C-EnterNet team is utilizing all approaches to refine future source attribution efforts.

TABLE 12.1
C-EnterNet plan and achievements with regards to Source Attribution in 2009

Approach / Objective	Data used	Status*	Main results/ conclusions	Main output
1. MICROBIAL SUBTYPING				
1.a Informal, descriptive comparison of subtyping data for various pathogens between the humans and the potential sources.	Annual subtyping data (e.g., serotypes, phagetypes, PFGE patterns) obtained through C-EnterNet's active food, animals, and water surveillance and the enhanced human surveillance in Sentinel Site 1.	D Each year	-Travel- and non travel-related human cases do differ in terms of subtyping (e.g., Salmonella sero and phagetypes). - overall, the match between subtypes seen in human cases and those observed in sources is weak to limited.	2006, 2007, 2008, and 2009 C-EnterNet's Annual Reports (particularly the section Exposure Sources in the 2007 Annual Report).
1.b Adaptation of the 'Danish *Salmonella* source account' model to the Canadian data.	Published sero- and phagetyping data from NML for the human side and sero- and phagetyping from LFZ and CFIA for the source side. Data between 2003 and 2007.	I	Data analysis planned for second half of 2010.	Expected publication in 2011.
2. QUANTITATIVE MICROBIAL RISK ASSESSMENT				
2.a QMRA of cryptosporidiosis related to recreational water.	Data collected through the C-EnterNet's active water surveillance in Sentinel Site 1 from March 2005 to Dec 2007, plus extra data from literature or other data sources.	D	See the published paper.	Pintar, Fazil, Pollari, Charron, Waltner-Toews, McEwen. "A risk assessment model to evaluate the role of fecal contamination in recreational water on the incidence of cryptosporidiosis at the community level in Ontario". Risk Analysis. 2010; Jan;30(1):49-64
2.b QMRA of cryptosporidiosis related to municipally treated water.	Data collected through the C-EnterNet's active water surveillance in Sentinel Site 1 from March 2005 to Dec 2007, plus data from the episodic survey on water consumption habits conducted by C-EnterNet in Sentinel Site 1, plus extra data from literature or other data sources.	D	See the published paper.	Pintar et al. "Assessing the risk of infection by Cryptosporidium via consumption of municipally treated drinking from a surface water source in a South-western Ontario Community". Submitted for peer-reviewed publication.
3. RISK EXPOSURE ASSESSMENT				
3.a *Campylobacter* risk exposure assessment.	Data of detection and quantity of *Campylobacter* in retail meat, food animals and water collected through C-EnterNet in Sentinel Site 1, plus extra data collected in the same area from other sources.	P	Planned for 2011.	—
4. OUTBREAK DATA ANALYSIS				
4.a Descriptive analysis of foodborne outbreak data from all over the world with comparison between large geographical regions.	4,093 reports of foodborne outbreaks that have occurred worldwide between 1998 and 2007. They were compiled by the LFZ Food Safety and Risk Assessment group through a systematic scan on the internet.	D	See the published paper.	Greig and Ravel. "Analysis of foodborne outbreak data reported internationally for source attribution". International Journal of Food Microbiology. 2009, 130: 77-87.

4.b Descriptive analysis of Canadian foodborne outbreak data with an historical perspective.	Reports of Canadian foodborne outbreaks combining 3 data sets covering 30 years (1976-2005). The data sets were provided by the Bureau of Microbial Hazards, Health Canada, the Center for Foodborne, Environmental, and Zoonotic Infectious Diseases and the Laboratory for Foodborne Zoonoses, both with the Public Health Agency of Canada.	D	See the published paper.	Ravel, Greig, et al. "Estimating Human Gastrointestinal Illness Attribution in Canada through Foodborne Outbreak Data Analysis". Journal of Food Protection, 2009, 72(9): 1963-1976
5. CASE-CONTROL STUDIES				
5.a Enteric disease case-control study.	Risk factors of enteric disease cases over a 12 month period as collected through the enhanced human surveillance in C-EnterNet Sentinel Site 1 plus risk factors for controls enrolled in the same area, over the same period of time, through an episodic study undertaken by C-EnterNet through a contract.	I P	Data collection for the healthy control group between August 2009 and July 2010. Data analysis in late 2010.	Publication expected for 2011.
5. b General case-case comparison.	Risk factors data of human enteric disease cases collected yearly through C-EnterNet in Sentinel Site 1.	D each year	Relative risk factors for each enteric disease pointing out some specific potential sources (no formal testing).	2006, 2007, 2008, and 2009 C-EnterNet Annual Reports.
5.c Specific case-case comparison for cryptosporidiosis.	Risk factors data of human enteric disease cases collected from April 2005 to December 2007 through C-EnterNet in Sentinel Site 1.	D	See the published paper.	Pintar, Pollari, Waltner-Toews, Charron, McEwen, Fazil, Nesbitt. "A modified case-control study of cryptosporidiosis (using non-Cryptosporidium infected enteric cases as controls) in a Southwestern, Ontario community". Epidemiology & Infection, 2009, 137 (12): 1798-1799.
5.d Epidemiological and microbial description of travel-related cases compared to the domestically-acquired enteric infections.	Risk factors data collected yearly through C-EnterNet's enhanced human surveillance in Sentinel Site 1. Risk factors collected through C-EnterNet's enhanced human surveillance in Sentinel Site 1 from June 2005 to May 2009.	D	The travel-related cases can represent an important proportion (up to 50% or more) of all cases depending on pathogens and years. See the Results section below (Result #1).	2006, 2007, 2008, and 2009 C-EnterNet Annual Reports. Ravel, Nesbitt, Marshall, Sittler, Pollari. "Description and burden of travel-related cases caused by enteropathogens reported in a Canadian community". Journal of Travel Medicine, Fall 2010.
6. INTERVENTION STUDY				
Considered when designing C-EnterNet's surveillance system. Feasible only once several sentinel sites in operation.	—	—	—	—

7. EXPERT ELICITATION				
7. Food safety expert elicitation survey	Survey conducted in fall 2008 according to a methodology developed and used in the USA. A list of 150 food safety experts was built according to a snow-ball approach. The experts were from various fields (e.g., public health, government, food safety, university, industry) and were all located in Canada. Sixty-six experts responded to the survey.	D	See the results section below (Result #2)	Ravel, Davidson, Ruzante, Fazil. "Foodborne proportion of gastrointestinal illness: Estimates from a Canadian expert elicitation survey". Foodborne Pathogens and Disease.
		I	Analyses of the expert opinion on the proportion attributed to each food category in progress.	A second publication expected in 2011.
8. MISCELLANEOUS				
Seasonality in human salmonellosis and exposure sources.	*Salmonella* data collected through the C-EnterNet's enhanced lab-based human surveillance and its active retail surveillance in Sentinel Site 1 from June 2005 to May 2009. Analyzed for seasonal pattern in human incidence, potential risk factors, and contamination in exposure sources.	D	See the section below (Result #3).	Ravel, Smolina, Sargeant, Cook, Marshall, Fleury, Pollari. "Seasonality in Human Salmonellosis: Assessment of Human Activities and Chicken Contamination as Driving Factors". Foodborne Pathogens and Disease July 2010, 7(7): 785-794.
Seasonality in human campylobacteriosis and exposure sources	*Campylobacter* data collected through the C-EnterNet's enhanced lab-based human surveillance and its active retail surveillance in Sentinel Site 1 from June 2005 to May 2009. Analyzed for seasonal pattern in human incidence, potential risk factors, and contamination in exposure sources.	I	Analysis in progress.	Publication expected in 2011.

* D= done; I= in progress; P= planned

Information generated

Result #1: Description and burden of travel-related cases caused by enteropathogens reported in a Canadian community.

Summary:
Risk of infections by enteropathogens among individuals traveling outside their country of residence is considered important. Such travel-related cases (TRC) have been poorly estimated and described in Canada.

Data from an enhanced, passive surveillance system of diseases caused by enteropathogens within a Canadian community from June 2005 to May 2009 were used to describe TRC in terms of disease (pathogen, symptoms, hospitalization, duration and timing of sickness relative to return), demographics (age, gender) and travel (destination, length, accommodation), and to compare them with non-TRC.

Among 1,773 reported cases, 446 (25%) were classified TRC, with 9% of them being new immigrants. The main TRC diseases were campylobacteriosis, salmonellosis and giardiasis. Disease onset occurred before return in 42% of TRC. Main destinations were Latin America/ Caribbean and Asia. No differences by month and year were observed for onset, departure and return dates. In addition to new immigrants, three subgroups of TRC based on travel destination, length of travel, type of accommodation, and age were identified and some diseases were more frequently observed in these subgroups. Generally, TRC did not differ from domestic cases in terms of age, gender, symptoms, hospitalization, and disease duration. *Campylobacter coli* and *Salmonella* Enteritidis were significantly more frequent among TRC.

TRC of diseases caused by enteropathogens that are reportable in Canada represent a significant proportion of the burden of the total diseases. Subgroups of TRC exist and are associated with certain diseases. These results help inform the assessment of the actual risk related to travel for each subgroup of travellers and quantify the attribution of travelling abroad to the overall burden of these gastrointestinal diseases.

Reference: Ravel, Nesbitt, Marshall, Sittler, Pollari. "Description and burden of travel-related cases caused by enteropathogens reported in a Canadian community". Journal of Travel Medicine. 2011, 18(1);8-19

Result #2: Foodborne proportion of gastrointestinal illness: Estimates from a Canadian expert elicitation survey

Summary:
The study used a structured expert elicitation survey to derive estimates of the foodborne attributable proportion for nine illnesses caused by enteric pathogens in Canada. It was based on a similar study conducted in the United States and focussed on *Campylobacter*, *Escherichia coli* O157:H7, *Listeria monocytogenes*, non typhoidal *Salmonella enterica*, *Shigella*, *Vibrio*, *Yersinia enterocolitica*, *Cryptosporidium parvum*, and Norwalk-like virus. For each pathogen, experts were asked to provide their best estimate and low and high limits for the proportion of foodborne illness relative to total cases. In addition, they provided background information with regards to food safety experience, including self-evaluated expertise for each pathogen on a 5-point scale. A snowball approach was used to identify 152 experts within Canada. The experts' background details were summarized using descriptive statistics. Factor analysis was used to determine whether

the variability in best estimates was related to self-assessed level of expertise or other background information. Cluster analysis followed by beta function fitting was undertaken on best estimates from experts who self-evaluated their expertise 3 or higher. In parallel, Monte Carlo resampling was run using triangular distributions based on each expert's best estimate and its limits. Sixty-six experts encompassing various academic backgrounds, fields of expertise and experiences relevant to food safety provided usable data. Considerable variation between experts in their estimated foodborne attributable proportions was observed over all diseases, without any relationship to the expert's background. Uncertainty about their estimate (measured by the low and high limits) varied between experts and between pathogens as well. Both cluster analysis and Monte Carlo resampling clearly indicated disagreement between experts for *Campylobacter*, *E. coli* O157, *L. monocytogenes*, *Salmonella*, *Vibrio*, and *Y. enterocolitica*. In the absence of more reliable estimates, the observed discrepancy between experts must be explored and understood before one can judge which opinion is the best.

Reference: Ravel, Davidson, Ruzante, Fazil. "Foodborne proportion of gastrointestinal illness: Estimates from a Canadian expert elicitation survey". Foodborne Pathogens and Disease. 2010, 7(12);1463-1472.

Result #3: Seasonality in Human Salmonellosis: Assessment of Human Activities and Chicken Contamination as Driving Factors

Summary:
This study used integrated surveillance data to assess the seasonality in retail chicken contamination and of human activities and their role on the seasonality of human endemic salmonellosis. From June 2005 to May 2008, reported cases of salmonellosis were followed-up comprehensively using a standardized questionnaire while 616 retail chicken breasts were systematically tested for *Salmonella,* in one Canadian community. Poisson regression was used to model seasonality of human cases, *Salmonella* in retail chicken and to assess the relationship between these and selected meteorological variables. The case-case approach was used to compare the activities of salmonellosis cases that occurred during the summer peak to the other cases. There were 216 human endemic salmonellosis cases (incidence rate: 14.7 cases / 100,000 person-years), predominantly of Typhimurium and Enteritidis serotypes (28.4 and 20.8%, respectively). The monthly distribution of cases was associated with ambient temperature (p<0.001) with a significant seasonal peak in June (p=0.03) and July (p=0.0005), but it was not associated with precipitation (p=0.38). Several activities reported by cases tended to be more frequent during summer. Particularly, attending a barbeque and gardening within the 3 days prior to the disease onset were two significant risk factors for salmonellosis in June or July compared to the salmonellosis cases that occurred in the other months. Out of all chicken samples, 185 (30%) tested positive for *Salmonella* spp., Kentucky being the dominant serotype (44.3% of positive samples). The monthly proportion of positive chicken samples showed no seasonal variations (p=0.30) and was not associated with the monthly count of human cases (p=0.99). In conclusion, even though evidence generally supports chicken as a primary vehicle of *Salmonella* to humans, the contamination of retail chicken was not driving the seasonality in human salmonellosis. Attending a barbeque or gardening during the hotter months of the year should be further assessed for their risk.

Reference: Ravel, Smolina, Sargeant, Cook, Marshall, Fleury, Pollari. "Seasonality in Human Salmonellosis: Assessment of Human Activities and Chicken Contamination as Driving Factors". Foodborne Pathogens and Disease. July 2010, 7(7): 785-794.

Component	Sample Type	Speciation Or Microscopic ID	Molecular ID	Enumeration (MPN)	Serotyping	Phagetyping	Ribotyping	AMR	PFGE	Genotyping
RETAIL MEAT	Skin-off chicken breasts Ground beef Pork chops	**Continuous:** *Salmonella* *Campylobacter* *Yersinia* (pork) *VTEC* *Listeria*		*Salmonella,* *Campylobacter* *Yersinia*	*Salmonella* *Yersinia*	*Salmonella*	*Listeria*		*Salmonella* *Listeria*	
RETAIL PRODUCE	Bagged leafy greens	**Episodic:** *Salmonella* *Campylobacter* *Verotoxigenic E. coli* *Generic E. coli* *Listeria monocytogenes* *Cryptosporidium* *Giardia* *Cyclospora*	**Episodic:** *Shigella* Norovirus Rotavirus		*Salmonella* *Listeria* Verotoxigenic *E. coli*	*Salmonella*	*Listeria*		*Salmonella* *Listeria* Verotoxigenic *E. coli*	*Cryptosporidium* *Giardia* *Cyclospora* Norovirus Rotavirus
ON-FARM	Fresh and stored pooled manure (dairy, beef, swine broiler chickens)	**Continuous:** *Salmonella* *Campylobacter* *Yersinia* (swine) *E. coli O157:H7*			*Salmonella* *Listeria* *Yersinia*	*Salmonella*	*Listeria*		*Salmonella* *E. coli O157:H7*	
WATER	Raw surface water	*Salmonella* *Campylobacter* *Yersinia* *E. coli O157:H7* *Giardia* *Cryptosporidium*			*Salmonella* *Yersinia*	*Salmonella*			*Salmonella* *E. coli O157:H7*	*Cryptosporidium* *Giardia*
HUMAN	Human stool samples	*Salmonella* *Campylobacter* *Yersinia* *E. coli O157:H7* *Cryptosporidium* *Giardia* *Shigella* *Listeria*			*Salmonella* *Listeria* *Yersinia*	*Salmonella*		*Salmonella* *Campylobacter*	*Salmonella* *E. coli O157:H7*	

APPENDIX B: Questionnaire Results

	Campylobacteriosis		Salmonellosis		*E. coli* O157:H7		Yersiniosis		Giardiasis		Cryptosporidiosis		Amoebiasis		All
Case Information															
	Cases	Non-cases[b]	Cases	Non-cases[b]	Cases	Non-cases[b]	Cases	Non-cases[b]	Cases	Non-cases[b]	Cases	Non-cases[b]	Cases	Non-cases[b]	Cases
Total number endemic cases[a]	99	170	82	187	10	258	7	262	40	229	17	252	13	256	268
Number with exposure data	80	150	75	155	10	219	6	224	33	197	13	217	12	218	229
Proportion with exposure data	81.0	88.0	91.0	83.0	100.0	85.0	86.0	85.0	83.0	86.0	76.0	86.0	92.0	85.0	85.0
Exposure Information															
Private well - main water source	11	16	16	14	20	14	0	15	18	14	23	14	8	15	12
Municipal - main water source	66	53	55	59	50	58	50	58	55	58	54	58	42	58	53
Drank untreated water	8	5	2	9	0	7	0	7	14	5	0	7	33	6	14
Swam	18	19	16	20	10	19	20	19	30	17	36	18	0	19	28
in a lake	9	6	5	8	0	7	0	7	9	7	15	6	0	7	5
in a pool	8	9	8	8	10	8	17	8	9	8	15	8	0	9	13
in a river	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
Drank unpasteurized milk	4	3	1	4	10	3	0	3	0	4	15	2	0	3	3
Ate undercooked food	11	6	10	6	0	8	0	8	0	9	0	8	0	8	9
Attended a barbecue	26	19	15	26	50	20	0	22	25	22	27	22	0	23	19
Ate in a restaurant	28	22	28	22	40	23	17	24	3	27	31	23	8	24	24
Ate meat from butcher shop	6	5	4	6	20	5	17	5	6	6	0	6	0	6	4
Ate meat from private kill	0	2	1	1	10	1	0	1	0	2	8	1	0	1	1
Shopped at butcher shop	8	13	5	14	40	9	40	10	10	11	18	11	40	10	9
Contact with household pet	58	48	58	48	50	52	20	52	37	53	42	52	14	53	45
cats	23	16	20	18	10	19	17	18	15	19	15	19	0	19	13
dogs	45	29	35	34	40	34	0	35	18	37	38	34	8	35	26
reptile	1	5	9	1	0	4	0	4	0	4	0	4	0	4	2
Visited farm animal areas	16	11	4	18	30	12	0	14	14	13	45	11	0	14	13
cats	1	0	0	1	0	0	0	0	0	1	0	0	0	0	0
dogs	4	0	0	2	0	1	0	1	0	2	0	1	0	1	1
horses	0	2	0	2	0	1	0	1	6	1	8	1	0	1	1
cattle	4	3	0	5	20	3	0	4	3	4	15	3	0	4	2
pigs	1	1	0	2	10	1	0	1	3	1	0	1	0	1	1
poultry	5	1	0	4	0	3	0	3	3	3	8	2	0	3	2
Lived on a farm/rural	14	17	15	16	20	15	0	16	14	16	42	14	0	16	14
On-farm animal exposures															
cats	3	1	1	2	0	2	0	2	0	2	8	1	0	2	1
dogs	3	3	4	2	0	3	0	3	0	3	8	2	0	3	2
horses	4	1	3	2	0	2	0	2	0	3	0	2	0	2	2
cattle	4	3	1	5	0	4	0	4	3	4	15	3	0	4	2
pigs	0	2	1	1	10	1	0	1	0	2	8	1	0	1	1
poultry	4	3	1	4	0	3	0	3	3	3	15	2	0	3	2
sheep	1	1	0	2	10	1	0	1	0	2	8	1	0	1	1

Note: Potential exposures are highlighted in yellow when the percentage for the specific disease is at least 5% greater than the exposure for the other enteric diseases combined.

[a] Does not include Cyclosporiasis, Hepatitis A, Listeriosis, or Shigellosis.

[b] Non-cases include all other enteric cases with exposure information.

APPENDIX C: Method Changes in 2009

Retail Sampling

In April 2009, C-EnterNet's routine retail sampling program expanded to include the testing of bagged, pre-washed, ready-to-eat leafy greens, including lettuce, and spinach. Each week 14 samples were collected from 4 to 5 randomly selected grocery stores within Sentinel Site 1 for a total of 376 samples in 2009. Samples were shipped to the Bureau of Microbial Hazards, Health Canada for pathogen testing by molecular (and microscopy on PCR-positive samples for parasites) and culture methods.

Method Change

In March 2009 (water program) and June 2009 (retail program, pork) the sensitivity of the *Yersinia* culture method was increased by the laboratory, by incorporating the addition of a KOH/NaCl solution to the broth. This additional step in the laboratory protocol resulted in a significant improvement in the isolation of *Yersinia* from water and raw pork chop samples.

APPENDIX D: Retail Enumeration Results

	# Samples Tested for Presence/ Absence	# Positive Samples by Presence/ Absence	MPN/g of sample				
			Below Detection (< 0.3)	0.3-10	11-100	101-1000	>1000
Campylobacter							
Pork	200	1	1				
Chicken	200	92	69	22		1	
Beef	200	1					
Salmonella							
Pork	200	3	3				
Chicken	200	57	52	3	2		
Beef	200	1	1				
Listeria							
Pork	163	16	11	2	3		
Chicken	165	31	21	7	3		
Beef	164	20	15	4	1		
Yersinia							
Pork	200	60*	37	6	2	5	4

* MPN/g results were not available for 6 pork samples

APPENDIX E: Sources Cited

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NOTES