

Appendix A

**ENTEROHEMORRHAGIC *ESCHERICHIA COLI*  
INFECTION (EHEC)**

**A RISK PROFILE**

**AUGUST 2002**

## **1. PATHOGEN-FOOD COMMODITY COMBINATION(S) OF CONCERN**

### ***Escherichia coli***

*E. coli* strains that are pathogenic for humans and cause diarrheal illness may be categorized into specific groups based on virulence properties, mechanisms of pathogenicity, and clinical syndromes. These categories include enteropathogenic *E. coli* (EPEC), enterotoxigenic *E. coli* (ETEC), enteroinvasive *E. coli* (EIEC), diffusely-adherent *E. coli* (DAEC), enteroaggregative *E. coli* (EaggEC), and enterohemorrhagic *E. coli* (EHEC). The EHEC group comprises a subset of Shiga toxin-producing *E. coli* (STEC),<sup>8</sup> which include strains of *E. coli* that cause bloody diarrhea in many infected patients. Shiga toxin-producing *E. coli* strains produce either or both of two phage-encoded toxins, Shiga toxin 1 (Stx1) and Shiga toxin 2 (Stx2). However, Stx production alone may not be enough to cause illness. Some EHEC strains also contain genes that encode for the ability to attach to and damage intestinal tract cells, causing what is commonly referred to as attaching-and-effacing lesions. *E. coli* O157:H7 is the single most important EHEC serotype in relation to public health. For a detailed review of the pathogenesis of EHEC and other STEC, interested readers are referred to recent publications by Paton and Paton (1998) and Nataro and Kaper (1998).

#### **1.1 Pathogen of concern**

Enterohemorrhagic *Escherichia coli* (EHEC) were first identified as human pathogens in 1982, when *E. coli* strains of a previously uncommon serotype, O157:H7, were implicated in two outbreaks of hemorrhagic colitis (bloody diarrhea) in the United States (U.S.). Since then, outbreaks of this new pathogen have become a serious public health problem throughout many regions of the world (Schlundt 2001; Clarke et al. 2002). The continued occurrence of large outbreaks and an increase in the incidence of reported cases suggests *E. coli* O157:H7 is an emerging pathogen (Tauxe 1997; Altekruze et al. 1997). Also in the 1990s, EHEC strains of other serogroups such as O26, O103, O111, and O145 were increasingly linked to human illness as illustrated by surveillance data from Japan (Table 1). This is also suggested by the WHO, which reported that O26, O103, O111 and O145 are the most important non-O157 serogroups (WHO 1998). Three outbreaks in the U.S. have been ascribed to non-O157 EHEC: a family outbreak of *E. coli* O111 with a case of HUS, a milk-associated episode of *E. coli* O104:H21 affecting 18 individuals and an outbreak of gastrointestinal illness, including bloody diarrhea, associated with *E. coli* O111:H8 in 56 persons (CDC 2000). Non-O157 serotypes of *E. coli* including O26:H11, O111:H8, O103:H2, O113:H21, and O104:H21 have been responsible for a small number of outbreaks in other parts of the world (CDC 1995b; Goldwater and Bettelheim 1995; Paton et al. 1996; Robins-Browne et al. 1998). In a cluster of three cases of HUS caused by O113:H21 in Australia, this organism was found not to have the attaching-and-effacing gene (Paton et al. 1999).

Table 1: Serotypes of human EHEC isolates from 1999-2000 in Japan\*

Serotype	1999 Cases (% of total)	2000 Cases (% of total)
O157	1394 (72.1)	1158 (69.9)
O26	346 (17.9)	377 (22.8)

<sup>8</sup> STEC are also referred to as VTEC (verotoxigenic *Escherichia coli*) in some member states. Both names are frequently employed in the scientific literature.

O111	81 (4.2)	42 (2.5)
All other	112 (5.8)	79 (4.8)

\*<http://idsc.nih.gov/iasr/22/256/graph/t2563.gif>

Most clinical laboratories do not routinely screen for non-O157 EHEC, because of the lack of a biochemical marker (Mead et al. 1998) and consequently there is little surveillance conducted for these EHEC infections. While *E. coli* O157:H7 are easily differentiated biochemically from other enteric *E. coli* because they ferment sorbitol slowly, diagnostic methods for identifying non-O157 EHEC are not widely available in most laboratories; consequently infections caused by these pathogens are often not confirmed. Recently, new methods for the detection of O103, O111, O26 and O145 serogroups have been developed; these advances may facilitate the collection of more data regarding the prevalence and significance of these serotypes as it pertains to human foodborne illness (Cudjoe 2001). Mead et al. (1999) has estimated that the incidence of non-O157 EHEC is between 20% and 50% that of *E. coli* O157:H7 infection. Because *E. coli* O157:H7 is the single most important EHEC serotype in relation to public health and because of the current paucity of epidemiologic data for non-O157 EHEC, this risk profile will emphasize *E. coli* O157:H7.

## 1.2 Commodities of Concern

In order to choose the most appropriate product to consider in this risk profile, the frequency with which various products were implicated in causing *E. coli* O157:H7 infection was considered. To accomplish this, we evaluated available studies of sporadic cases of *E. coli* O157:H7 infection and outbreak investigation reports. Sporadic cases account for the majority of reported cases in a given year and therefore may be more representative of persons with *E. coli* O157:H7 infection. For example, 75% of reported cases in one region of the U.S. during 1991-97, and 83% of reported cases in another region during 1992-1999, were sporadic (OCD 1998, Proctor and Davis 2000). Food vehicles implicated most frequently are raw or inadequately cooked foods of bovine origin, especially undercooked hamburgers and unpasteurized milk; however, an increasing number of outbreaks are associated with the consumption of raw or minimally processed fruits and vegetables.

- Enterohemorrhagic *Escherichia coli* (EHEC) were first identified as human pathogens in 1982, when *E. coli* strains of a previously uncommon serotype, O157:H7, were implicated in two outbreaks of hemorrhagic colitis (bloody diarrhea) in the United States.
- Since then, outbreaks of this new pathogen have become a serious public health problem throughout the industrialized world and have led to the designation of *E. coli* O157:H7 as an emerging pathogen.
- EHEC strains of other serogroups such as O26, O103, O111, and O145 have been increasingly linked to human illness.
- At present, *E. coli* O157:H7 is the single most virulent and important EHEC serotype in relation to public health and since epidemiologic data for non-O157 EHEC is lacking, CCFH should limit its immediate efforts to *E. coli* O157:H7.

### Foods of bovine origin

Case control studies of sporadic illness have described the association between ground beef consumption (in most cases, undercooked product) and *E. coli* O157:H7 infection (Table 2). Grinding meat introduces the pathogen into the interior of the

meat. When ground beef does not achieve the required internal temperature (e.g., > 68°C)<sup>9</sup> or when the product is cooked unevenly, *E. coli* O157:H7 may survive. In most countries, many thousands of pounds of meat trim from many carcasses are ground together; therefore, a small number of carcasses with *E. coli* O157:H7 can contaminate a large supply of ground beef. Additionally, contaminated beef may transfer *E. coli* O157:H7 to meat grinding equipment, which may later contaminate other lots of raw meat. Ground-beef products, therefore, pose a greater hazard than intact cuts of meat. Dry fermented meats have also been implicated in reported outbreaks of EHEC infection (Tilden et al. 1996). A case-control study showed a relation between consumption of two sausages, mortadella (cooked) and teewurst (fermented, containing beef), and illness (Ammon et al. 1999).

Table 2: Case-control studies implicating ground beef in *E. coli* O157:H7 infection

Study Reference	Study Type	Finding
Slutsker 1998	Case-control, sporadic illness	Consumption of round beef with "pink center" had 34% population attributable risk.
Mead 1997	Case-control, sporadic illness	45% of ill persons consumed ground beef with "pink center" in the preceding week while only 33% of controls did the same.
Kassenborg 2001	Case-control, sporadic illness	Ground beef with "pink center" was a statistically significant risk factor while consumption of just ground beef was not.
MacDonald 1988	Prospective study	Rare ground beef was consumed more often by ill persons than healthy persons.
Le Saux 1993	Case-control, sporadic illness	Consumption of undercooked ground beef had an attributable risk factor of 17%.

Outbreak investigations have also contributed significantly to our understanding of how *E. coli* O157:H7 is transmitted. Ground beef was identified as the transmission source in seven out of 13 (53.9%) outbreaks that occurred between 1982 and 1993 in the U.S. (Griffin 1995); however, outbreaks have been attributed to foodborne, waterborne and person-to-person means of transmission.

Beef was cited as the source of 46% of the foodborne outbreaks with a known vehicle of transmission in the U.S. between 1993-1999. Of the 21 beef-associated outbreaks that occurred during 1998-1999, ground beef was identified as the vehicle in 19 (Table 3). Five (26.3%) of the 19 ground beef/hamburger-associated outbreaks occurred in multiple states. Two outbreaks in 1999 were attributed to roast beef and one of these was a result of environmental contamination from manure in a pasture where a picnic was held.

<sup>9</sup> Recommendations ranging between 68.3 and 71 °C have been made. In some cases these are associated with holding times at the specified temperature such as 15 seconds.

Table 3: Food Vehicles Implicated in Outbreaks of *E. coli* O157:H7, U.S., 1998–1999

Vehicle	1998	1999	Total
Ground beef/hamburger	10	9	19
Roast beef	0	2	2
Combined green leafy vegetables	4	7	9
Lettuce	1	3	4
Coleslaw	2	1	3
Salad	1	1	2
Milk	2	0	2
Other (none with greater than 1 total)	5	5	10
Total	21	21	42

Sources: CDC 1999b; CDC 2001c.

Other products of bovine origin that have been implicated in a number of outbreaks of *E. coli* O157:H7 infection include raw and improperly pasteurised cow's milk as demonstrated by a O104:H21 outbreak from contaminated milk (Feng et al. 2001). Milk borne outbreaks mostly have been associated with the consumption of raw milk or milk products from local farms. Raw milk is often contaminated with enteric organisms during its collection and may result in a direct risk for consumers choosing to drink raw milk. Effective pasteurisation eliminates pathogens from milk, including *E. coli* O157:H7.

#### Foods of non-bovine origin

A variety of foods may occasionally become contaminated with *E. coli* O157:H7 by cross-contamination with beef or other meats and contaminated kitchen surfaces during food preparation. Mayonnaise and mayonnaise-based dressings and sauces were identified as the most likely foods to have been contaminated in a series of outbreaks of *E. coli* O157:H7 infections in the U.S. (Jackson et al. 2000). Survival studies indicated that mayonnaise could serve as a vehicle for EHEC infections when stored at refrigeration temperatures, despite the low pH of mayonnaise. Sandwiches were cited as the probable source in an outbreak of *E. coli* O157:H7 at a nursing home (Carter et al. 1987). Additionally, several outbreaks associated with wild game meat have been reported (Asakura et al. 1998, Keene et al. 1997).

Fruits and vegetables contaminated with *E. coli* O157:H7 have accounted for a growing number of recognised outbreaks (Table 3). Examples of vegetables, fruits, and sprouts that have been implicated in foodborne outbreaks of *E. coli* O157:H7 infection include fresh potatoes (Morgan 1988), lettuce (Ackers et al. 1998, Mermin et al. 1997, Hilborn et al. 1999), radish (Michino et al. 1998), alfalfa sprouts (Breuer et al. 2001, MMWR 1997a) and cantaloupe (Del Rosario and Beuchat 1995). As a whole, leafy green vegetables were cited as the source of 26% of the foodborne outbreaks with a known vehicle of transmission in the U.S. between 1998-99. Contamination of vegetables may occur in several ways, however, the use of manure or water contaminated with fecal matter is one possible route (Solomon et al. 2002; Wachtel et al. 2002; Solomon et al. 2002b). In a number of the instances cited above, manure from nearby cattle lots was suspected to be the original source of *E. coli* O157:H7 (Ackers et al. 1998; Hilborn et al. 1999). Similarly, in an accidental release of tertiary-treated sewage that had not been treated with chlorine, cabbage plants were found to have *E. coli* strains (not containing stx1, stx2 or eae genes) associated with the plant roots when control fields did not (Wachtel et al. 2002). Another means of contamination of these products is cross-contamination in the retail, or consumer kitchen between contaminated meat products and produce.

Fruit juices have also been implicated in outbreaks of *E. coli* O157:H7 infection (Besser 1993, CDC 1996, CDC 1997, Cody 1999, MMWR 1997b, Steele 1982). Although the low pH of fruit juices will generally not allow the survival and outgrowth of many of the Enterobacteriaceae, these products may allow survival of *E. coli* O157:H7 when they become contaminated because of the microorganism's high acid-tolerance. Although the exact mechanisms of contamination for these outbreaks were not clearly determined, animal manure was suspected to have contaminated the fruit.

In summary, there are many foodborne pathways by which individuals can be exposed to *E. coli* O157:H7. Other significant risk factors reported in literature are exposure to farm animals or the farm environment, eating at a table service restaurant, using immune suppressive medication (for adults only) and obtaining beef through a private slaughter arrangement (Kassenborg et al. 2001; OCD 1998). Current data based on both outbreaks and sporadic infections indicate that consumption of ground beef is still the single most important source of foodborne *E. coli* O157:H7 illness. Leafy green vegetables are the second most significant cause of human foodborne illness cases of *E. coli* O157:H7 as they are subject to contamination and they are eaten raw. Table 5 in the final section of this document describes the national and academic risk assessments that have been conducted to date for *E. coli* O157:H7 infection.

Due to their relevance to human cases of *E. coli* O157:H7 infection, ground beef and green leafy vegetables contaminated by *E. coli* O157:H7 from bovine faeces are the focus of this risk profile. Commodities worthy of future consideration include raw milk products, unpasteurized cider and sprouted seeds.

- Foods associated either directly or indirectly with animals (meat or dairy products) or foods subject to contamination by animal waste products such as fertilizer or agricultural runoff are frequently implicated as vehicles of transmission for human illness.
- Beef was cited as the source of 46% of the foodborne outbreaks with a known vehicle of transmission in the U.S. between 1993-1999.
- Leafy green vegetables were cited as the source of 26% of the foodborne outbreaks with a known vehicle of transmission in the U.S. between 1998-99.
- Ground-beef products pose a greater hazard than intact cuts of meat.
- For these reasons, this working group will only address the threat associated with ground beef and leafy green vegetables that have been contaminated with bovine faeces as vehicles of transmission.

## **2. Description of the public health problem**

### **Epidemiology**

Following ingestion of *E. coli* O157:H7, the human response ranges from asymptomatic infection to death. To cause disease after ingestion, the *E. coli* O157:H7 must survive acidic conditions within the stomach prior to moving to distal portions of the gastrointestinal tract. Disease due to *E. coli* O157:H7 occurs primarily in the colon. The incubation period from the time of ingestion to the first symptoms ranges from one to eight days. Asymptomatic shedding of *E. coli* O157:H7 has been documented (Swerdlow 1997); however, the proportion of exposed individuals who shed *E. coli* O157:H7 but do not

develop symptoms is unknown. Typically the illness begins with abdominal cramps and nonbloody diarrhea that can, but does not necessarily, progress to bloody diarrhea within two to three days (Griffin 1995, Mead et al. 1998). Usually 70% or more of symptomatic patients will develop bloody diarrhea; however, as many as 95% have been observed in other studies (Ostroff et al. 1989; Bell et al. 1994). More severe manifestations of *E. coli* O157:H7 infection include hemorrhagic colitis (grossly bloody diarrhea), hemolytic uremic syndrome (HUS)<sup>10</sup> and occasionally thrombotic thrombocytopenic purpura (TTP).

Symptoms of hemorrhagic colitis include severe abdominal cramps followed by grossly bloody diarrhea and edema (swelling), erosion, or hemorrhage of the mucosal lining of the colon (Su and Brandt 1995). Hemorrhagic colitis may be the only manifestation of *E. coli* O157:H7 infection, or it may precede development of HUS. Complications from hemorrhagic colitis associated with *E. coli* O157:H7 include upper-gastrointestinal bleeding and stroke (Su and Brandt 1995). Roberts et al. (1998, citing Boyce et al. 1995a, Ryan et al. 1986) estimates the mortality rate of those suffering hemorrhagic colitis without progression to HUS to be 1%, although Griffin (personal communication) believes this rate is too high. Approximately 30% to 45% of patients are hospitalized (Ostroff et al. 1989, Le Saux et al. 1993, Bell et al. 1994, Slutsker et al. 1998). Of the 631 cases reported to FoodNet sites in 1999, 39% were hospitalized (CDC 2000b). Treatment for the more serious manifestations of *E. coli* O157:H7 infection is supportive and the use of antimicrobial agents has been debated (Mead 1998).

The incidence of *E. coli* O157:H7 infection varies by age group, with the highest incidence of reported cases occurring in children. In addition to children, elderly are known to be susceptible to *E. coli* O157:H7 infection. A report detailing a Scottish outbreak resulting from contaminated beef involving at least 292 confirmed cases of *E. coli* O157:H7 infection resulted in 151 hospitalizations and 18 deaths; all fatalities were elderly patients (Ahiied 1997).

The number of reported *E. coli* O157:H7 cases derived from surveillance is known to underreport the true disease burden. Underestimation of the actual incidence of infection occurs for a variety of reasons, including:

- some infected persons do not seek medical care;
- physicians do not perform diagnostic testing on all patients with symptoms of infection;
- some persons who obtain medical care do not provide a stool specimen;
- laboratories do not culture all stool samples for *E. coli* O157:H7. In a 1994 national survey in the U.S., 70 (54.3%) of 129 randomly selected clinical laboratories reported that they did not routinely test all stools or all bloody stools for *E. coli* O157:H7 (Boyce 1995b).
- some proportion of laboratory results are false negatives; and
- not all culture-confirmed infections are reported by health care providers and laboratories to public health authorities.

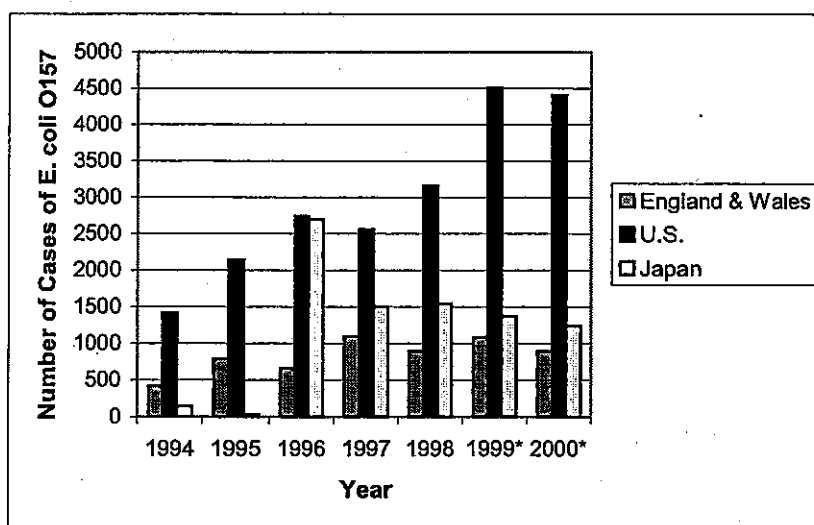
Using surveillance data, and accounting for the factors that contribute to underreporting, Mead (1999) estimated that 73,480 cases of *E. coli* O157:H7 infection occur annually in the U.S. and that 85% (62,456 cases) are a result of foodborne exposure.

*E. coli* O157:H7 was designated by the Council of State and Territorial Epidemiologists as a nationally notifiable disease in the U.S. beginning in 1994. During 1994-2000, the number of reported cases of *E.*

<sup>10</sup> a combination of renal failure, low platelet counts and hemolytic anemia

*coli* O157:H7 in the U.S. increased more than two-fold from 1,420 (0.8/100,000 people) in 1994 to 4410 (approximately 1.6/100,000 people) in 2000 (CDC 1999, CDC 2001) (Figure 1). Cases in the U.S. are reported by passive surveillance through the National Notifiable Diseases Surveillance System (NNDSS).<sup>11</sup> This is a passive surveillance system in which health care providers report notifiable disease cases to local or state health departments. Other national or regional surveillance systems include (1) Enter-net<sup>12</sup> which includes a catchment area of 15 European Union (EU) member states as well as Switzerland and Norway, (2) The Communicable Disease Network Australia – National Notifiable Surveillance System,<sup>13</sup> (3) Japan's Statistics on Communicable Diseases in Japan (former Ministry of Health and Welfare) and the National Epidemiological Surveillance of Infectious Diseases (NESID) which are reported in Infectious Agents Surveillance Reports<sup>14</sup> and (4) the EU's Zoonoses Reporting System.<sup>15</sup> In addition to these surveillance systems, the EU, Japan and U.S. have each developed a pulsed-field gel electrophoresis (PFGE) database to assist in epidemiological investigations of disease from this and other bacteria. The increase in reported cases of *E. coli* O157:H7 over time is probably due to a combination of factors, including (1) improvement in the effectiveness of the surveillance system over time, (2) increased awareness of *E. coli* O157:H7 infection among health care providers and the public leading to improved detection and reporting, (3) enhanced ability to detect disease through better diagnostic tests, and (4) a true increase in the incidence of disease. Figure 1 illustrates the rising incidence of *E. coli* O157:H7 infection in three different regions of the world. For more information on the global impact of EHEC see the WHO Consultation report (WHO 1997).

**Figure 1.** Number of reported cases of *E. coli* O157:H7 infection, U.S. (1994-2000),<sup>a</sup> England and Wales (1994-2000),<sup>b</sup> and Japan (1996-2000);<sup>c</sup> \*provisional data are presented from 1999 and 2000 for the US.



<sup>11</sup> <http://www.cste.org/nndss/reportingrequirements.htm>

<sup>12</sup> [http://www.phls.org.uk/topics\\_az/ecoli/data.htm](http://www.phls.org.uk/topics_az/ecoli/data.htm)

<sup>13</sup> <http://www.health.gov.au/pubhlth/cdi/nndss/year054.htm>

<sup>14</sup> <http://idsc.nih.go.jp/iasr/22/256/tpc256.html>; <http://idsc.nih.go.jp/index.html>; Note that in the former system, known as the Ministry of Health and Welfare, communicable diseases in Japan were reported in "Statistics on Communicable Diseases in Japan" and, during a transitional period, in the "Annual Report on National Epidemiological Surveillance of Infectious Diseases". The new system, known as the National Epidemiological Surveillance of Infectious Diseases (NESID), publishes "Infectious Agents Surveillance Reports" monthly describing pathogen isolates and related information and "Infectious Disease Surveillance Data" annually describing notified human cases (IDSD is currently only available by CD-ROM format).

<sup>15</sup> EU Council Directive 92/117/EEC; [http://europa.eu.int/eur-lex/en/com/pdf/2001/en\\_501PC0452\\_01.pdf](http://europa.eu.int/eur-lex/en/com/pdf/2001/en_501PC0452_01.pdf)



- a) CDC, NNDSS; Cases include suspect and confirmed human isolations.
- b) PHLS Laboratory of Enteric Pathogens; Cases include only isolates, obtained from stool samples, that are submitted to PHLS from laboratories in England and Wales. They are confirmed, serotyped, phage typed and VT typed at PHLS.
- c) Ministry of Health and Welfare, National Epidemiological Surveillance of Infectious Diseases; Cases are restricted to those with stool samples that have been culture confirmed and include all O157 serotypes.

In 1996, the Emerging Infection Program Foodborne Diseases Active Surveillance Network (FoodNet) began a program of active surveillance of clinical laboratories for specific foodborne diseases, including *E. coli* O157:H7. Five states in the U.S. participated initially (Minnesota, Oregon, selected counties of California, Connecticut and Georgia) (CDC 2001a). As of 2000, the areas under active surveillance included 8 states representing 29.5 million persons (10.8% of the 1999 U.S. population). The number of cases of *E. coli* O157:H7 infection reported annually to FoodNet ranged from 388 in 1996 to 631 in 2000 (Bender et al. 2000, CDC 2000b, CDC 2001a). Because the population under surveillance has increased, it is more appropriate to compare the number of reported cases per 100,000 persons in a population.

Data on the prevalence of symptomatic *E. coli* O157:H7 infection prior to the inception of FoodNet are scarce and include studies which estimate between two and 10 cases for every 100,000 persons (Ostroff et al. 1989, MacDonald et al. 1988). The higher estimates obtained in some of these studies is likely a consequence of the active method for data collection and may provide a more accurate estimate of the incidence of *E. coli* O157:H7 infection thereby suggesting that statewide passive surveillance programs are hindered by underreporting.

HUS is the most common cause of acute renal failure in young children, yet it also has long-term complications. In Belgium 97% of HUS cases in 2000 were associated with *E. coli* O157:H7 infection (Pierard et al. 1997). Siegler (1994) found that HUS causes chronic renal sequelae, usually mild; in 51% of survivors (48% of all cases) however, Elliot et al. (2001) has observed significantly lower renal failure statistics in Australia. Neurologic complications occur in about 25% of HUS patients (Mead et al. 1998). Generally neurologic symptoms are mild, but serious complications, such as seizure, stroke and coma, can occur (Su and Brandt 1995). Similar to treatment for *E. coli* O157:H7 infection, only symptomatic treatment for neurologic complications is available, making this manifestation of HUS especially dangerous and an important cause of death in HUS patients. Other complications of HUS include pancreatitis, diabetes mellitus and pleural and pericardial effusions (Mead et al. 1998). In a nationwide study of HUS patients, 46 (55%) of 83 patients required either peritoneal dialysis or hemodialysis during the acute phase of their illness (Banatvala et al. 2001). Siegler et al. (1994) found that severe kidney or neurological impairments (end stage renal disease or stroke) occurred in 9 (5.7%) of 157 HUS cases over a 20-year period in Utah. A number of studies have suggested the mortality rate associated with HUS is between 3 and 7% (Martin 1990; Tarr 1987; Rowe 1991; Mahon 1997; Banatvala 2001; Siegler 1994).

The percent of *E. coli* O157:H7 infections which progress to HUS varies between sporadic cases and those associated with outbreaks. Between 3% to 7% of sporadic, and 20% or more of outbreak associated cases of *E. coli* O157:H7 infection will progress to HUS (Mead 1998). The proportion of patients who develop HUS following *E. coli* O157:H7 infection is influenced by a variety of factors including age, bloody diarrhea, fever, elevated leukocyte count, and toxin type (Griffin et al. 1995). Wong (2000) found that 10 (14.1%) of 71 children with *E. coli* O157:H7 infection developed HUS. Similarly, the severity of HUS illness may differ between sporadic cases and those associated with outbreaks; outbreaks often resulted in a shorter diarrheal prodrome, a higher rate of bloody diarrhea and severe hemorrhagic colitis (Elliot et al. 2001).

Between 1997 and 1999 at FoodNet sites located within the U.S., the overall incidence of HUS among children younger than 15 years of age was 0.7 per 100,000; this is similar to the frequency observed in other nations such as Austria (0.65 per 100,000) and Australia (0.64 per 100,000) (Elliot et al. 2001). For children younger than 5, the incidence was 1.4 and 1.35 per 100,000 in the U.S. and Australia respectively (CDC 2000b). In a nationwide study of 83 patients with HUS in the U.S., 46 (55.4%) were younger than 5 years old and an additional 27 (32.5%) were 5 to 17 years old (Banatvala et al. 2001). In 1999, 35.3% of reported HUS cases in the U.S. occurred in 1- to 10-year-olds, 17.6% of cases occurred in 10- to 20-year-olds, and 14.1% of cases occurred in persons older than 60 (CDC 2000b). Similarly, analyses of HUS incidence in Belgium found the majority (35/46) of HUS cases were in children (Pierard et al. 1997). The overall findings of this study demonstrated that the burden of illness from HUS is comparable between Australia, North America and Europe. A national study of postdiarrheal HUS in the U.S. estimated that  $\leq 20\%$  of HUS cases were due to non-O157 EHEC; however, the authors qualified that estimate, commenting that it was difficult to determine the proportion of EHEC-associated HUS due to non-O157 EHEC (Banatvala et al. 2001). In Australia, between July 1994 and June 1998, only 8% of the EHEC associated cases of HUS were the result of *E. coli* O157 infection (Elliot et al. 2001). This suggests that while illness from HUS is similar on different continents, the predominant EHEC serotype responsible may vary.

Occasionally, patients with *E. coli* O157:H7 are diagnosed as having thrombotic thrombocytopenic purpura (TTP), a condition similar to HUS but more likely to occur in adults and with more prominent neurological findings and less renal involvement. In the study by Banatvala et al. (2001), of 73 children and 10 adults that met the case definition of HUS, 8 (11.0%) children and 8 (80.0%) adults also met the case definition for TTP. None of the 8 children, but 2 (25.0%) adults died. There are many causes of TTP other than the association with *E. coli* O157:H7 and prior to the 1980s, gastrointestinal infections had not been strongly implicated in the pathogenesis of TTP (CDC 1986). When associated with *E. coli* O157:H7 infection, TTP is probably the same disorder as HUS (Mead et al. 1998).

- Between 70 and 95% of symptomatic patients develop bloody diarrhea.
- More severe manifestations of *E. coli* O157:H7 infection include hemorrhagic colitis (grossly bloody diarrhea), HUS and occasionally TTP.
- The incidence of *E. coli* O157:H7 infection varies by age group, with the highest incidence of reported cases occurring in children.
- In addition to children, elderly are known to be susceptible to *E. coli* O157:H7 infection.
- A number of national and regional disease surveillance systems exist that record *E. coli* O157 infection.
- While the incidence of hemolytic uremic syndrome is similar on different continents, the predominant EHEC serotype responsible may vary.

### **3. Food production, processing, distribution and consumption**

#### **1. The farm to table continuum**

As was previously mentioned, EHEC strains including *E. coli* O157:H7, have been isolated from the faeces or gastrointestinal tract of cattle, sheep, horses, pigs, turkeys, dogs, and a variety of wild animal species (Kudva 1996; Rice and Hancock 1995; Hancock et al. 1998b; Heuvelink et al. 1999); consequently, foods associated either directly or indirectly with animals (meat or dairy products) or foods subject to contamination by animal waste products (for instance as manure fertilizers) are frequently implicated as vehicles of transmission for human illness. Epidemiological studies have found that cattle manure is the primary source of most human *E. coli* O157:H7 infections. In fact, *E. coli* O157:H7 has been described as

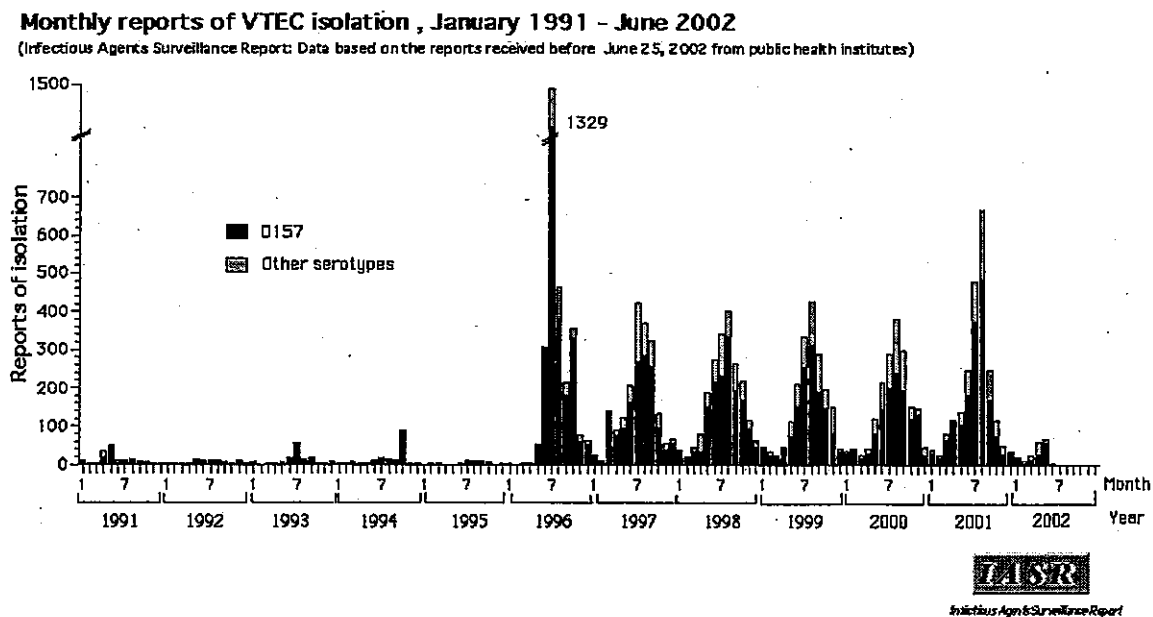
“ubiquitous” in dairy and beef cattle and is present at least occasionally on most farms or feedlots (Hancock et al. 1998a; Hancock et al. 2001). Factors contributing to the presence of *E. coli* O157:H7 include:

- the pathogen’s ability to survive for at least 4 months in water trough sediments (Hancock et al. 1998a); and
- the pathogen’s presence in some animal feeds (Hancock et al. 2001).

Many of the risk factors that are thought to influence *E. coli* O157:H7 prevalence and their levels in cattle apply to whole herds rather than to individual cattle. Therefore, mitigation strategies typically target herd-level risk factors for *E. coli* O157:H7 control. The roles that water, including treated or untreated sewage used to irrigate animal feed, and feed itself play in colonizing herds may prove to be critical to on farm management strategies and should be considered (Anderson et al. 2001, Hancock et al. 2001, LeJeune et al. 2001). The type of herd does appear to have an affect on the prevalence of *E. coli*. Herds of feedlot cattle such as steers and heifers are more likely to have colonized animals than breeding herds, which are comprised of cows and bulls. Additionally, when a feedlot herd is positive it is likely to have significantly more colonized animals than breeding cattle herds (USDA, 2001). Limited evidence suggests that dairy and cow-calf herds are similar with respect to *E. coli* O157:H7 (Lag Reid et al. 1999; Sargeant et al. 2000).

An increased seasonal incidence of *E. coli* O157:H7 infections in cattle and human populations has been demonstrated in the warmer months of the year (Hancock et al. 1997a, 1997b; Griffin 1998; Van Donkersgoed et al. 1997, Heuvelink et al. 1998); this is evidenced in data from Japan (Figure 2). It is therefore not surprising that the incidence of HUS is also more common in the summer months ([www.hcsc.gc.ca/hpb/lcdc/bmb/epiic95/95\\_ii\\_e.html](http://www.hcsc.gc.ca/hpb/lcdc/bmb/epiic95/95_ii_e.html); Mead and Griffin 1998; CDC 1999a; CDC 2000b, Van de Kar 1996; Van Duynhoven accepted for publication). Of the sporadic cases of human *E. coli* O157 infection reported by FoodNet sites, 70% occurred during June through September for the years 1996 to 1998 (Bender et al. 2000; CDC 1999a). Similarly, *E. coli* O157 outbreaks also occur more frequently in the summer (CDC 1999a; CDC 2000c; CDC 2001c).

Figure 2: From IASR Infectious Agents Surveillance Report at <http://idsc.nih.gov/prompt/graph/vt9.gif>



*E. coli* O157:H7 has also been isolated from other water sources such as ponds, streams and wells (Wang and Doyle 1998 Hancock et al. 1998a Kudva et al. 1998; Sargeant 2000). Tainted water used to

clean food or as an ingredient in a food commodity (e.g. juice) can also contribute to the contamination of foodstuffs.

### Characteristics of the commodities

Leafy green vegetables grown low to the ground are a recognized cause of *E. coli* O157:H7 outbreaks. Contamination of vegetables may occur in several ways, however, the use of manure or water contaminated with fecal matter is one possible route (Solomon et al. 2002; Wachtel et al. 2002; Solomon et al. 2002b; Wachtel et al. 2002b). When contaminated, the fact that fresh produce is minimally processed and consumed raw increases the likelihood of *E. coli* O157:H7 infection. The source of *E. coli* O157:H7 contaminated manure can be, and frequently is, of bovine origin. Mitigation strategies that control *E. coli* O157:H7 on farm will likely impact the level of contamination of produce.

Meat becomes contaminated with *E. coli* O157:H7 when beef carcasses come into contact with contaminated hides and faeces during the slaughter process (Elder et al. 2000). A determination of the quantitative association between the incoming status of cattle and the outgoing status of harvested meat is critical in an exposure assessment. This quantitative correlation between pre-harvest and post-harvest contamination is best predicted using fecal *E. coli* O157:H7 prevalence data (Elder et al. 2000).

A number of factors have a significant influence on the survival and growth of *E. coli* O157:H7 in food, including temperature, pH, salt, and water activity (Meng and Doyle 1998). Studies on the thermal sensitivity of *E. coli* O157:H7 in ground beef have revealed that the pathogen has no unusual resistance to heat and that heating ground beef sufficiently to kill typical strains of *Salmonella* will also kill *E. coli* O157:H7. The optimal temperature for growth of *E. coli* O157:H7 is approximately 37°C (98.6°F), and the organism will not grow at temperatures below 8°C to 10°C (46°F to 50°F) or above 44°C to 45°C (Doyle and Schoeni 1984; Buchanan and Doyle 1997). *E. coli* O157:H7 survives freezing, with some decline in the concentration of *E. coli* O157:H7 (Ansary et al. 1999).

*E. coli* O157:H7 has been reported to be more acid resistant than other *E. coli*. Acid resistance enhances the survival of *E. coli* O157:H7 in mildly acidic foods and may explain its ability to survive passage through the stomach and cause infection at low doses. The ability to be acid resistant varies among strains and is influenced by growth phase and other environmental factors. Once induced, acid resistance is maintained for long periods of time during cold storage (Meng and Doyle 1998). Stationary-phase *E. coli* O157:H7 are more resistant than growing cells to acid (Meng and Doyle 1998). The presence of other environmental stresses, such as temperature or water activity stress will raise the minimum pH for growth (Buchanan and Doyle 1997). *E. coli* O157:H7 survives in such foods as dry salami, apple cider, and mayonnaise, which were previously considered too acidic to support the survival of foodborne pathogens. Published literature contains conflicting reports about the efficacy of acid spray washing of beef carcasses. A study by Brachett et al. (1994) found that warm and hot acid sprays did not significantly reduce the concentration of *E. coli* O157:H7 on beef carcasses; however, two recent studies have found organic acids to be effective in reducing the presence of *E. coli* O157:H7 on beef carcasses (Berry and Cutter 2000; Castillo et al. 2001). These apparently contradictory results may reflect differences in acid resistance among strains of *E. coli* O157:H7 (Berry and Cutter 2000).

*E. coli* O157:H7 can survive for extended periods under conditions of reduced water activity while refrigerated; however, the organism does not tolerate high salt conditions (Buchanan and Doyle 1997).

### Retail and Consumer Behavior

The food preparation industry and consumer choices and behaviors have a large influence on the probability of contracting an *E. coli* O157:H7 infection. Specifically, inadequately cooked beef (in particular ground or minced products) is correlated to infection as previously discussed. Cooking these products to an internal temperature of  $\geq 68^{\circ}\text{C}$  has been shown to be an adequate precaution; however, consumers continue to choose undercooked beef products.<sup>16</sup> It has also to be borne in mind that in

<sup>16</sup> <http://www.fsis.usda.gov/oa/news/1998/colorpr.htm>

certain countries consumption of products made from raw minced beef (such as “tartare” steak, beef “americaine”) is common. Similarly, awareness of and precautions against cross-contamination between raw meat products and either cooked foods or raw vegetables would limit the likelihood of infection.<sup>17</sup> Consumer behavior that can limit illness from vegetables contaminated at the farm is likely limited to thoroughly cleaning produce, in particular commodities to be consumed raw. In the case of sprouted seeds and some fruits and vegetables (Solomon et al. 2002), which have been shown to integrate the bacteria, washing may not be a sufficient intervention.

2.

### 3. Interventions

A Canadian risk assessment was published that predicted the reduction in illnesses to be expected from various mitigation scenarios for ground beef (Table 4) (Cassin et al. 1998). These mitigations included achievement of maximum temperature control during storage, preslaughter screening of cattle faeces and cooking at appropriate temperatures. Based on the Cassin approach, an Australian risk assessment also modeled risk mitigation scenarios including hot water decontamination of carcasses, irradiation of frozen boxed beef, preslaughter reduction in faecal concentrations, retail temperature control and consumer education about good cooking practices (Lammerding et al. 1999; P Vanderlinde, personal communication).

- Meat becomes contaminated with *E. coli* O157:H7 when beef carcasses come into contact with contaminated hides and faeces during the slaughter process.
- An increased seasonal incidence of *E. coli* O157:H7 infections in cattle and human populations has been demonstrated in the warmer months of the year.
- *E. coli* O157:H7 has no unusual resistance to heat and heating ground beef sufficiently to kill typical strains of *Salmonella* will also kill *E. coli* O157:H7.
- Consumer choices and actions such as undercooking beef have a large influence on the probability of contracting an *E. coli* O157:H7 infection.

Due to the impact on-farm cattle colonization can have on other commodities such as leafy green vegetables, interventions that control *E. coli* O157:H7 in live animals are also of great interest. These include the impact of employing probiotic (Zhao 1998) bacterial flora in cattle, the impact of various feeding regimens (Lung et al. 2001), the result of different composting protocols (Cray et al. 1998) and the impact of various irrigation methods.

## **4. Risk Assessment Needs and Questions for the Risk Assessors**

**Is a microbiological risk assessment appropriate to fulfill the desired CCFH output(s)?**

As both leafy green vegetables grown low to the ground contaminated by bovine faeces and ground beef are commonly associated with these illnesses they should each be considered, either together or separately, in a risk assessment. It would be useful for CCFH to request a risk assessment be conducted to specifically assess the impact of any proposed guidance. These same documents also suggest useful questions to be directed to the risk assessors to evaluate methods for minimizing (1) the colonization of cattle with *E. coli* O157:H7, (2) the contamination of

<sup>17</sup> [http://www.fsis.usda.gov/oa/pubs/keep\\_apart.htm](http://www.fsis.usda.gov/oa/pubs/keep_apart.htm)

ground beef with faeces and (3) the contamination of produce with bovine faeces. It is possible that the benefits achievable through downstream interventions are less important than those obtained via interventions at the farm due to the multiple infection pathways (and commodities) that can be minimized through effective manure management.

Any risk assessment undertaken by FAO/WHO should provide an estimate of the risk of illness and death from *E. coli* O157:H7 in countries where sufficient data are available. Illness must be explicitly defined by CCFH as a specified endpoint. Also, an assessment should be provided of the potential benefit (i.e. number of cases of illness and death prevented) for intervention strategies including those currently in effect or under consideration in member states [listed in brackets] such as:

**Measure effect of controlling *E. coli* O157:H7 at the farm level with respect to subsequent agricultural use of manure.**

- The impact of employing probiotic bacterial flora in cattle.
- The impact of various feeding regimens.
- Outcomes from different composting protocols
- The impact of various irrigation methods (Solomon et al. 2002).
- The impact of hygienic measures at the farm (such as cleaning and disinfection of premises between lots)
- Calf management at weaning

**Measures to minimize fecal shedding of *E. coli* O157:H7 in animals presented for slaughter [Belgium, Sweden, others]**

- Enforcing or tightening controls on animal feed quality.
- The impact of employing probiotic bacterial flora in cattle.
- The impact of various feeding regimens and fasting before transport
- Calf management at weaning
- Monitoring of human illness with mandatory testing of farms linked to an outbreak of human illness resulting from *E. coli* O157:H7. Restrictions on positive farms include controls over the sale of live animals, and restriction on the sale of animals for slaughter (slaughter hygiene and swab tests at slaughter).

**Measures to minimize contamination of carcasses at slaughter [Australia, Denmark, Germany, Portugal, Sweden, U.K., U.S.].**

- Scoring the level of fecal contamination on the hides of incoming animals with remedial action (e.g. logistic slaughter) when scores exceed a predetermined level (clean cattle policy).
- Rodding and clipping the esophagus and bagging and tying the rectum.
- Hygienic dressing and evisceration.
- Random testing of *E. coli* O157:H7 on beef carcasses in the slaughterhouse. A positive test results in testing on the farm of origin. Positive on-farm tests resulting in increased farm sanitation measures.
- Random testing of carcasses for (generic) *E. coli* contamination and measures to improve sanitation when predetermined levels are exceeded.
- Random testing of carcasses for enterobacteriaceae.
- Visual inspections of carcasses.
- The use of HACCP in slaughter and processing.

- Different decontamination procedures.

#### **MEASURES TO MINIMIZE CONSUMER EXPOSURE TO CONTAMINATED PRODUCTS** [Australia, U.K., U.S.].

- Random testing for *E. coli* O157:H7 in trim and ground meat.
- Random testing for (generic) *E. coli* or enterobacteriaceae in ground beef.
- Destruction or diversion of positive product to cooked product.
- Irradiation or pasteurization.
- Specified cooling guidance.
- Enhanced hygienic practice during cutting, boning and other steps between slaughter and retail.
- Restaurant cooking requirements.
- HACCP in the food production and service sector.

#### **Retail Codes / Consumer education.**

- Cooking ground beef to a specified internal temperature as indicated by the use of a meat thermometer [Canada, Germany, U.S.]

#### **Measures to minimize contamination of food products in international trade** [special consideration of CCFH]

- A certification program to insure that exported products meet the acceptable level of protection of the importing country.

A WHO Consultation discussed the global impact of EHEC and the controls and prevention strategies employed by a number of nations (WHO 1997). European national guidance documents and mitigations are discussed in the European Commission Health and Consumer Protection Directorate-General's report SANCO/4320/2001.<sup>18</sup> Canada has produced (1) interim guidelines for the control of verotoxigenic (Shiga toxin producing) *Escherichia coli* including *E. coli* O157:H7 in ready-to-eat fermented sausages containing beef or a beef product as an ingredient,<sup>19</sup> (2) policies concerning raw products of animal origin,<sup>20</sup> and (3) policies for ground beef containing *E. coli* O157:H7.<sup>21</sup> Australia has developed microbiological standards and Advisory Guidelines for the Hygienic Production of Uncooked Fermented Comminuted Meat Products based on generic *E. coli* so as to include all STEC serotypes.<sup>22</sup>

These issues will need to be prioritized by the Committee and explicit questions for the risk assessors subsequently formulated.

### **5. Available Information**

A number of countries have evaluated the risk associated with foodborne *E. coli* O157. These assessments are listed in Table 5. Specifically, Canada has analyzed the risk associated with *E. coli* O157:H7 infection

<sup>18</sup> [http://europa.eu.int/comm/food/fs/inspections/special\\_reports/sr\\_rep\\_4320-2001\\_en.pdf](http://europa.eu.int/comm/food/fs/inspections/special_reports/sr_rep_4320-2001_en.pdf)

<sup>19</sup> [http://www.hc-sc.gc.ca/food-aliment/english/organization/microbial\\_hazards/guideline\\_for\\_fermented\\_sausages.html](http://www.hc-sc.gc.ca/food-aliment/english/organization/microbial_hazards/guideline_for_fermented_sausages.html)

<sup>20</sup> [http://www.hc-sc.gc.ca/food-aliment/english/organization/microbial\\_hazards/pdf/rfao\\_sept21.pdf](http://www.hc-sc.gc.ca/food-aliment/english/organization/microbial_hazards/pdf/rfao_sept21.pdf)

<sup>21</sup> [http://www.hc-sc.gc.ca/food-aliment/english/organization/microbial\\_hazards/guidelines\\_for\\_raw\\_ground\\_beef.html](http://www.hc-sc.gc.ca/food-aliment/english/organization/microbial_hazards/guidelines_for_raw_ground_beef.html)

<sup>22</sup> <http://www.anzfa.gov.au/foodstandards/oldfoodstandardscodecontents/partmeatcannedmeatandproductsthereof/c1meatgamemeatandrel686.cfm>

from consuming ground beef hamburgers (Cassin et al. 1998), sprouts (personal communication with Health Canada, January 2002) and juices (personal communication with Health Canada, January 2002) since each of these commodities have contributed to outbreaks or sporadic incidents of illness in that nation. An academic group in Canada has also assessed risk factors associated with on-farm *E. coli* O157 prevalence in cattle (Jordan et al. 1999a, 1999b). The Netherlands chose to investigate steak tartare as the vehicle of transmission in their risk assessment because: (1) a steak tartare is thicker than a hamburger, therefore the risk of insufficient heating of the center is larger, (2) people tend to accept a partially raw tartare but do not accept a partially raw hamburger, (3) tartare is sometimes consumed raw (e.g., a tartare roll in snack bars). Steak tartare was chosen after also considering consumption levels. The U.S. has developed a farm-to-table risk assessment for *E. coli* O157:H7 in ground beef in addition to a comparative risk assessment for *E. coli* O157:H7 in tenderized and non-tenderized steaks. Due to the smaller contribution O157 serotypes make to human illness in Australia, that state has developed one risk assessment for O157:STEC and another for all STEC in ground beef production and in fermented meat.

- FAO/WHO may find many of these risk assessments useful in the development of a risk assessment for Codex. Further evaluation of each is necessary.



Table 5: Risk assessments for *E. coli* O157:H7

Nation	Topic	Reference
Australia	Ground Beef <sup>1</sup>	Lammerding 1999
Australia	STEC in Ground Beef <sup>1</sup>	Lammerding 1999
Canada	Ground Beef Hamburgers	Cassin 1998
Canada	Seeds/Beans and Sprouted Seeds/Beans <sup>2,3</sup>	Personal Communication with Health Canada
Canada	Unpasteurized Fruit Juice/Cider <sup>4</sup>	Personal Communication with Health Canada
Canada	Pre-harvest Husbandry Practices	Jordan 1999a,b
Ireland	Beef/Beef Products	<a href="http://www.science.ulst.ac.uk/food/E_coli_Risk_Assess..htm">www.science.ulst.ac.uk/food/E_coli_Risk_Assess..htm</a>
Netherlands	Raw Fermented Products	<a href="http://www.research.teagasc.ie/vteceurope/S+Gprog/hoornstrasg.html">www.research.teagasc.ie/vteceurope/S+Gprog/hoornstrasg.html</a>
Netherlands	Steak Tartare	RIVM report 257851003/2001
U.S.	Ground Beef <sup>5</sup>	<a href="http://www.fsis.usda.gov/OPPDE/rdad/FRPubs/00-023NReport.pdf">www.fsis.usda.gov/OPPDE/rdad/FRPubs/00-023NReport.pdf</a>
U.S.	Tenderized vs Non-tenderized Beef Steaks	Personal Communication with USDA

<sup>1</sup>ANZFA Food Standard Code 1.6.1 sets Microbiological limits for total generic *E. coli* in a variety of foods. <http://www.anzfa.gov.au/foodstandardscodecontents/standard16/index.cfm>. Additionally, dairy products must be produced from pasteurized milk.

<sup>2</sup>Subsequent policy and management documents include "Consultation/Policy Document: A Dialogue on Developing a Risk Management Strategy for Sprouted Seeds and Beans".

<sup>3</sup>Subsequent policy and management documents include "Code of Practice for the Hygienic Production of Sprouted Seeds"

<sup>4</sup>Subsequent policy and management documents include "Code of Practice for the Production and Distribution of Unpasteurized Apple and Other Fruit Juice/Cider in Canada"

<sup>5</sup>The U.S. has a microbiological criteria equivalent to a zero-tolerance for *E. coli* O157:H7 in raw ground beef

## 6. Data Gaps

Several data gaps have been identified based on currently available risk assessments for *E. coli* O157:H7, including:

- Information describing the human health impact of *E. coli* O157 in less developed nations.
- Commodities likely to be associated with *E. coli* O157 foodborne illness in less developed nations.
- Data regarding the exposure dose of *E. coli* O157:H7 likely to cause illness in susceptible populations.
- Frequency and severity of illness among children ages 0 to 5 from *E. coli* O157:H7 that become ill from consuming ground beef d in raw produce.

- Industry and consumer practices for various methods of cooking ground beef (e.g., grill vs. fry).
- Survival of *E. coli* O157:H7 on produce as a result of contamination by water or organic fertilizer.
- Information describing the critical contamination levels of meat products that may lead to cross contamination of uncooked produce.
- Information on the percentage of fresh leafy vegetables contaminated by bovine faeces containing *E. coli* O157:H7 as opposed to feral animal faeces, or human faeces
- Quantify the heat resistance (e.g., D and z values) of the individual strains of *E. coli* O157:H7 used in the Sporing (1999) study. Individual strains should be identified and characterized.
- Information on the maximum density of *E. coli* O157:H7 organisms in ground beef servings as a result of matrix effects, competitive microflora in ground beef, and environmental conditions (e.g., pH, water activity).
- Predictive microbiological data on the increase and decrease in the number of *E. coli* O157:H7 organisms in ground beef under various storage and preparation conditions along with frequencies of occurrence of these storage and preparation conditions.
- Data on cross-contamination of *E. coli* O157:H7 between carcasses during carcass splitting.
- Time-temperature data (quantitative) for chillers in slaughter establishments.
- Marketing data on the proportion of beef ground at slaughter versus at retail.
- Data on retail and consumer storage, cooking, and consumption (frequency and serving size) patterns by type of ground beef meal (e.g., grilled hamburger in July and baked meat loaf in October).
- Descriptive epidemiologic information about sporadic cases of *E. coli* O157:H7 illness, including the month of disease onset, age, sex, hospitalizations, summary of clinical manifestations including severe disease manifestations, and food vehicles involved (if known).
- Additional case-control studies of sporadic *E. coli* O157:H7 cases to calculate etiologic fraction attributable to ground beef.

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