**A review of *Escherichia coli* as an emerging food-borne pathogen**

**Why is it important?**

Hundreds of thousands of people are made ill by *Escherichia coli* (E. coli) each year, and hundreds of them die. In recent years, there has been an increase in outbreaks of Shiga toxin-producing *E. coli* (STEC), and thousands of sporadic cases of haemorrhagic colitis (bloody diarrhoea), some of which develop into the potentially fatal haemolytic-uraemic syndrome (HUS). These STEC outbreaks have had a significant impact on health care systems, agricultural production and trade in many countries around the world.

**What is *E. coli***?

*E. coli* is a bacterium that is commonly found in the gastrointestinal tract of humans and warm-blooded animals. Because of its high prevalence in the gastrointestinal tract and in faeces, *E. coli* is used as the preferred indicator of faecal contamination when assessing the safety of food and water. Most *E. coli* are harmless commensal organisms when contained in their natural intestinal habitat.

Different strains of *E. coli* are serious human gastrointestinal pathogens, and some are also pathogenic for young food production animals. Pathogenic *E. coli* are distinguished from other *E. coli* by their ability to cause illness through genetically controlled mechanisms such as toxin production, adhesion and invasion of host cells, interference with cell metabolism and tissue destruction.

*E. coli* have the ability to exchange genetic material via mobile genetic elements such as plasmids and bacteriophages, as an adaptation response to new and stressful environments. These genetic elements are believed to contribute to the emergence of pathogenic types with enhanced virulence, environmental survival and persistence in food systems.

**Pathogenic *E. coli* types and symptoms in humans**

Pathogenic *E. coli* are assigned to six groups or pathotypes, based on common mechanisms of pathogenicity and clinical syndromes: Shiga-toxigenic *E. coli* (STEC) or verotoxigenic *E. coli* (VTEC); enterohaemorrhagic *E. coli* (EHEC); enterotoxigenic *E. coli* (ETEC); enteroinvasive *E. coli* (EIEC); enteropathogenic *E. coli* (EPEC); enteroaggregative *E. coli* (EAggEC or EAEC); and diffusively adherent *E. coli* (DAEC).

Characteristics of the pathotypes are not exclusive and may be shared by more than one group. In general, the incubation period in human cases of *E. coli* disease ranges from three to eight days, with the appearance of a variety of gastrointestinal symptoms, ranging from mild to severe and bloody diarrhoea, mostly without fever.

Infected individuals and animals (with or without disease symptoms) can shed up to $10^6$ to $10^9$ colony-forming units (cfu) per gram of faeces.
The following are the main characteristics and distinctions among the six pathotypes:

- **STEC or VTEC** produce symptoms that range from mild to severe and bloody diarrhoea. STEC produce cytotoxins called verotoxins (VT) or Shiga toxins (Stx) (owing to their similarity to *Shigella dysenteriae* toxin). Up to 10 percent of cases can develop life-threatening HUS, particularly in young and elderly patients.

- **EHEC** are a subset of STEC typically associated with bloody diarrhoea and HUS. EHEC and EPEC produce intestinal epithelial cell changes called attaching and effacing lesions. STEC/EHEC are asymptptomatically carried by healthy animals such as cattle, sheep, goats and wildlife.

- **ETEC** commonly cause watery diarrhoea among infants and travellers to areas of the world with poor sanitation and hygiene. ETEC attach to the small intestine via colonization factor antigens and produce enterotoxins that are similar to *Vibrio cholerae* toxin and are either plasmid-mediated heat-stable toxins (ST) or chromosomally mediated heat-labile toxins (LT). These enterotoxins and their respective variants cause disruption of the sodium chloride balance in the intestine, resulting in profuse watery diarrhoea.

- **EIEC** penetrate and spread among intestinal cells causing extensive cell destruction resulting in mild to bloody diarrhoea similar to dysentery.

- **EPEC** cause profuse watery and sometimes bloody diarrhoea, particularly in infants in developing countries. EPEC adhere to the intestinal epithelium causing disruption of the cellular function. The pathology is associated with production of attaching and effacing lesions similar to those from EHEC. EPEC are distinct from STEC as they do not produce Stx.

- **EAggEC or EAEC** cause both acute and persistent watery and mucoid diarrhoea in young children. EAggEC attach to tissue culture cells in a distinctive aggregative pattern. A plasmid encoded enteroaggregative heat-stable toxin (EAST1) may contribute to diarrhoeal symptoms.

- **DAEC** are less well defined and cause diarrhoea in older children. DAEC are distinguished from EPEC and EAggEC by their diffuse adherence to tissue culture cells.

Ruminant animals, mainly cattle and wildlife, are recognized as the primary natural reservoir of STEC and, particularly, EHEC O157:H7. Pigs and poultry are not considered to be major sources of STEC for human infection in Europe.

Serotyping using antisera to somatic (O), flagella (H) and capsular (K) antigens is commonly used to distinguish *E. coli* strains, and there are now hundreds of antigenic types. Some pathotypes belong to certain serotypes, although this is not always exclusive. There are pathotype/serotype combinations more commonly associated with food-borne disease, such as EHEC belonging to the O157:H7 serotype. As not all strains are known to present a public health risk, it is important to distinguish pathogenic types based on pathotype as well as serotype.

**E. coli and food contamination**

Humans can acquire an infection with pathogenic strains through consumption of food and water directly contaminated with faeces or contaminated as a result of
cross-contamination from another food source. In addition there is possible contamination from direct human contact during food preparation. The epidemiology of food-borne pathogenic *E. coli* varies throughout the world. In communities with poor sanitation and hygiene, ETEC, EIEC and EPEC are prevalent. However, food-borne pathogenic *E. coli* have also emerged in communities with better developed sanitation and hygiene systems.

Food may also be contaminated and/or cross-contaminated during growth and harvest (horticulture products), collection (milk) or animal slaughter and carcass dressing (meat). Further contamination can occur during post-harvest handling, transport, processing and preparation.

*Fresh meat and raw milk* are considered common vehicles for *E. coli*, particularly EHEC O157:H7. Contamination of meat usually occurs during animal slaughter and carcass dressing as a result of poor hygiene practices and inadequate abattoir hygiene standards. Of particular importance are stages such as hide removal, evisceration and handling after dressing, which if not properly controlled are likely to result in contamination of meat by animal faeces.

*Fresh vegetables* can be contaminated with *E. coli* from animal and human faeces that may enter crop agro-ecosystems through inadequately composted manure, the use of untreated waste- and grey water for irrigation, contaminated seeds, wildlife and insect pests, and nematodes. Contaminated fresh produce that is eaten raw has become an emerging source of human *E. coli* infection. *E. coli* may survive in contaminated soil for up to 20 months. They can also survive for long periods on crop leaves and roots. Younger leaves tend to provide a better habitat than older ones, and leaves with higher levels of nitrogen or damaged leaves and fruits are able to support faster multiplication and prolonged survival of *E. coli*.

**Figure 1: STEC transmission**

![Figure 1: STEC transmission](Source: FAO)
Detection of pathogenic *E. coli* in foods

The wide diversity of *E. coli* pathotypes presents challenges for their detection. No single method can be used to detect all types, so methods target specific pathogenicity markers and serotypes. Detection of O157 EHEC has proved the easiest owing to their specific phenotype, virulence traits and serotype. Because even small numbers present in a food may constitute a health risk, enrichment is required to improve sensitivity to detection. Although genetic or immunological detection methods can be used to screen enriched samples, isolation and characterization of the bacterium are required for confirmation.

For the purposes of surveillance, outbreak investigation or assessment of health risk, pathogenic *E. coli* strains are usually typed according to a hierarchy of phenotype, pathotype, serotype, phagetype and DNA-based fingerprints (e.g., pulsed field gel electrophoresis).

The *E. coli* O104:H4 outbreak of 2011

On 26 May 2011, Germany reported what became the largest national outbreak of EHEC infections with the highest numbers of affected people developing HUS caused by Shiga-toxigenic *E. coli* infections. The outbreak was centred in northern Germany and peaked around 21 to 23 May 2011. It was officially considered over on 26 July 2011. During these two months, a total of 4,321 cases comprising 3,469 EHEC cases and 852 HUS cases were reported to the Robert Koch Institute (RKI) in Germany. In total 50 patients died. According to the European Centre for Disease Prevention and Control (ECDC), 76 EHEC cases, of which 49 developed HUS, including one patient who died (as of 22 July 2011) were reported across other European Union countries. All were linked to the German outbreak. Most of the patients who developed HUS were adults (89 percent, median age of 43 years) and women were overrepresented (68 percent).

All patients were infected with *E. coli* serotype O104:H4. This rare serotype was previously reported in few STEC and HUS cases, but never in foodstuffs. It was genetically characterized at the National Reference Laboratory for Salmonella and other Enteric Bacteria at RKI and found to possess characteristics of two types of pathogenic *E. coli* – EHEC and EAggEC. The organism was also resistant to many antimicrobials, and the combination of virulence factors suggested that the strain was more likely to be of human rather than animal origin.

The outcome of the epidemiological investigations in Germany suggested that the consumption of several types of sprouts was associated with the outbreak. The original source of the contamination was apparently traced to dry bean seeds used for sprouting.

On Friday 24 June, a cluster of 15 cases of HUS or bloody diarrhoea due to *E. coli* O104:H4 was identified in the Bordeaux area of France. The microbiological characteristics of the isolated strain of *E. coli* O104:H4 from three of the French HUS patients were similar to those of the isolated strain in the German outbreak, including the antibiotic resistance profile. A joint rapid risk assessment by the European Food Safety Authority and ECDC suggested that the consumption of fenugreek sprouts was the possible source of both the German and the French *E. coli* O104:H4 outbreaks.
Control of pathogenic *E. coli* in food and water

As the key points of control tend to vary with the specific pathotype implicated in an outbreak, knowledge of local food-borne disease epidemiology is essential in establishing an appropriate and effective control programme. This requires multidisciplinary approaches that address the interactions among humans, animals, plants and their ecosystems.

Control points along the food chain that will ensure the greatest reduction of risk to public health should be identified, and risk mitigation steps should be taken in accordance with recognized codes of good practice and relevant recommendations from veterinary and public health services. At the pre-harvest or pre-slaughter stage, such steps include minimizing pathogenic *E. coli* colonization of livestock – particularly ruminant – herds and prevention of manure contamination of crops. At the post-harvest or post-slaughter stage, they include slaughterhouse/milking shed hygiene and the application of good hygiene practices during carcase dressing, handling and packing of produce or meat.

Some *E. coli* strains can elicit stress responses that enhance their growth and persistence; for example, STEC may tolerate acid conditions in fruit juice and fermented meat and dairy products. *E. coli* are destroyed by thorough cooking, so any controlled heat treatment can be an effective means of elimination. The main challenges are therefore to prevent contamination or cross-contamination of foods that are to be eaten raw or with minimal processing, and to prevent post-process contamination of food.

Pre-harvest interventions in farm animal production

Strategies that reduce pathogen shedding in live animals can reduce pathogen populations in food animals before they enter the food chain. For example, abruptly switching cattle from a high-grain ration to a high-quality hay-based diet has been shown to reduce generic *E. coli* and *E. coli O157:H7* populations. The feeding of probiotic *Lactobacillus acidophilus* bacteria has also been shown to be effective and has been adopted for the pre-slaughter control of *E. coli O157:H7* in cattle. Further research is needed to elucidate the mechanism (e.g., competitive exclusion, physical removal, forage quality, tannins, lignin, other phenolics) by which forage feeding affects the microbial ecology of the bovine intestinal tract, including the ecology of *E. coli* and *E. coli O157:H7* populations, so that economically viable and practical dietary interventions can be implemented. Current areas of investigation include feed and water hygiene, dietary supplements and vaccination (a vaccine against *E. coli O157:H7* is commercially available). Research should also be aimed at improving understanding of the factors that cause individual animals to shed high numbers of pathogenic *E. coli* (so-called “super shedders”) and at identifying such animals and their farm holdings of origin. This would allow more risk-based controls to be applied to limit the risks of contamination from such animals or holdings.

Pre-harvest strategies in fresh produce and sprout production

Appropriate on-farm manure storage and handling practices that minimize runoffs from farms are important. Crop management can also reduce some of the factors...
associated with \textit{E. coli} populations and could reduce the risks of epidemics in humans. It is generally possible to reduce the survival and growth of \textit{E. coli} populations in crops by adopting good agricultural practices (FAO, 2011b). These include reducing the use of nitrogenous fertilizer, applying only treated or well-processed manure with a higher carbon-to-nitrogen ratio, applying compost, ensuring that seeds are not contaminated before planting, encouraging better animal and human hygiene in the field, and irrigating with clean water. While intended to reduce risks from \textit{E. coli}, these practices also support the sustainable intensification of crop production.

Low levels of pathogenic \textit{E. coli} grow prolifically during the production of sprouted seeds, so it is necessary to establish control to minimize initial seed contamination and limit subsequent growth. Guidance is available in the document CAC/RCP 53-2003 Annex for Sprout Production in FAO and WHO, 2007. This can be downloaded from the Web site\textsuperscript{1} or obtained on request from the Secretariat of the Codex Alimentarius Commission.\textsuperscript{2}

**Food processing and preparation**

Effective prevention of post-harvest contamination and cross-contamination can be achieved by applying practices based on the principles of good hygiene and manufacturing practices and Hazard Analysis Critical Control Point (HACCP)-based approaches. For the meat sector, the FAO (FAO and Fondation Internationale Carrefour, 2004) manual, \textit{Good practices for the meat industry}, outlines these principles. FAO is also involved in projects to strengthen veterinary public health systems and services

\textsuperscript{1} www.codexalimentarius.net/download/standards/10200/CXP_053e.pdf

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through veterinary supervision and inspection of animal slaughter establishments and practices, meat inspection and slaughterhouse hygiene.


Conclusions
A wide variety of pathogenic E. coli strains causing human food-borne diseases can be found in the gastrointestinal tract of animals and in the environment. Some animals or animal species can be asymptomatic carriers. E. coli strains are known for their propensity to exchange genetic elements and adapt to changes in their environment. Sometimes this leads to the emergence of strains with increased pathogenicity and survival capabilities.

The most effective way of preventing E. coli contamination of food and water is through the implementation of good hygiene and good practices at the primary production level and along the stages of the food supply chain, such as post-harvest/post-slaughter and any subsequent handling and preparation stage.

It is also necessary to strengthen systems for epidemiological surveillance of STEC, including non-O157 E. coli.

Resources


CAC. 2002. Risk profile for enterohaemorrhagic E. coli including the identification of the commodities of concern, including sprouts, ground beef and pork. CX/FH 03/5-Add.4 September 2002. Rome.


