

# *Escherichia coli* O157: H7 in Food With Health-Related Risks

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## ABSTRACT

*Escherichia coli* O157:H7 has been implicated in many cases of food contamination across the world, in both the beef and poultry industries, with approximately 1,000 *E. coli* O157 cases reported in UK each year. This includes 100 cases of haemolytic uraemic syndrome, a serious illness which can cause acute renal failure. Indeed, *E. coli* O157:H7 infections are of particular concern due to the potential severity of symptoms. An estimated 73,500 cases of illness, 2000 hospitalisations and 60 deaths occur each year in the USA due to *E. coli* O157 infection, costing approximately \$1 billion a year in medical costs and lost productivity. *E. coli* O157:H7 is regarded as being more transmissible than other *E. coli* serotypes for a number of reasons, including its increased tolerance to acid, which allows it to easily survive the acidic conditions of the stomach. This bacterium also produces Shiga toxins, which are heat stable, and therefore unaffected by conventional pasteurization methods. Small doses of fewer than 10 cells may lead to infection. Collectively, these factors make the control of *E. coli* O157:H7 an important issue in recent times for the food sector. Many preventative measures have been introduced and targeted at all stages of the food chain, from the farm, to the slaughterhouse, and to the preparation of food at home.

**Keywords-**Contamination; Food Safety; Pathogen; Microbiological Quality.

## INTRODUCTION

The increasing number and severity of food poisoning outbreaks on a global scale have considerably increased public awareness of food safety.<sup>1</sup> Well publicised cases of *Escherichia coli* (*E. coli*) serotype O157:H7 infections in particular are of concern due to the potential severity of symptoms.<sup>2</sup> Although *E. coli* O157:H7 was only first recognized as a cause of food borne illness just over 30 years ago<sup>3</sup>, it has been implicated in sporadic cases and large outbreaks of haemorrhagic colitis and fatal haemolytic uremic syndrome.<sup>4</sup> This serotype is regarded as being more transmissible than other *E. coli* serotypes due to a number of reasons, including its increased tolerance to acid, which allows it to easily survive the acid conditions of the stomach. This bacterium also produces Shiga toxins, which are heat stable, and therefore unaffected by conventional pasteurization methods.<sup>5</sup>

Small doses of fewer than 10 cells may lead to infection.<sup>3</sup> Collectively, these factors make the control of *E. coli* O157:H7 an important issue in recent times for the food sector.

The main causes for concern and product recalls associated with *E. coli* O157:H7 are meat products. In particular, cattle and sheep are major reservoirs for this pathogen<sup>6,7</sup> and contamination of carcasses and food products by animal faeces can lead to transmission of food borne pathogens to consumers.<sup>8</sup> Numerous interventions to be applied at the farm level have been investigated over

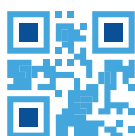
the past 20 years, but most have proven to be ineffective and/or impractical.<sup>9</sup> Furthermore, the conflict between demands for minimally processed foods and the modern requirement of long shelf-life and food safety is an issue for the food industry. This has led to interest in the use of natural antimicrobial products.

### *Clinical aspects of E. coli* O157:H7

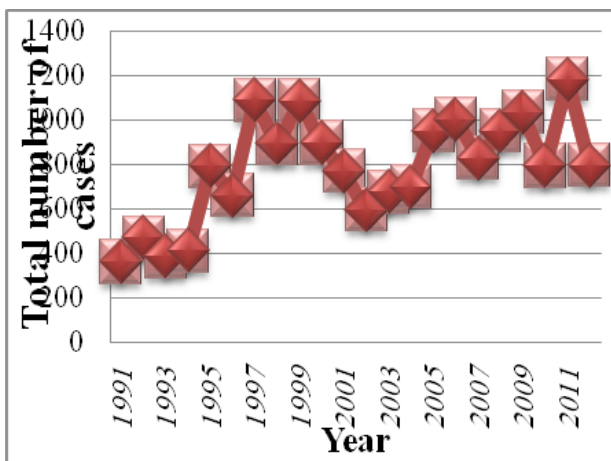
Infection with *E. coli* O157:H7 is asymptomatic in a large proportion of cases, but can also engender a wide range of clinical symptoms ranging from non-bloody diarrhoea to hemorrhagic colitis and other life-threatening complications. Serious health effects arising from infection can involve acute renal failure caused by haemolytic uraemic syndrome (HUS)<sup>10</sup>, and neurological problems in the form of thrombotic thrombocytopenic purpura (TTP).<sup>11,12</sup> Other rare complications include pancreatitis, diabetes mellitus, and pleural and pericardial effusions.<sup>13</sup> Occasionally, patients infected with *E. coli* O157:H7 suffer damage to their central nervous system as TTP, which typically includes seizures arising from hypertensive encephalopathy. Untreated TTP can have a mortality rate as high as 95%. Symptoms may include thrombocytopenia, fever, renal insufficiency, neurological deficit, microangiopathic haemolytic anaemia, headache, fatigue/malaise, altered mental status, and hemiplegia.<sup>10</sup>

### Epidemiology

Vero cytotoxin producing *Escherichia coli* VTEC has become the most frequently reported cause of bacteraemia



in England, Wales and Northern Ireland.<sup>14</sup> A report from<sup>2</sup> (Figure 1) suggests almost a 100% increase, from 595 to 1182, in the annual totals of VTEC infections in England and Wales between 2002 and 2011. To date, many parts of the world have witnessed outbreaks of VTEC infections involving serotype O157.<sup>11</sup> Infection rates differ widely between geographical regions. In Europe, Scotland possesses the highest infection rates with approximately 4 cases per 100,000<sup>11</sup>, while in Northern Europe infection rates are very low (e.g. 0.04 per 100,000 in Norway and Finland). In North America, the infection rate for *E. coli* O157:H7 was 0.9 per 100,000 in 2004. In Asia, Japan has experienced the most problems related to *E. coli* O157:H7 (2.74 per 100,000 averaged between 1999 and 2004).<sup>11</sup> An estimated 73,500 cases of illness, 2000 hospitalisations and 60 deaths occur each year in the USA due to *E. coli* O157 infection<sup>15</sup>, costing approximately \$1 billion a year in medical costs and lost productivity.<sup>16</sup> *E. coli* O157:H7 cases in England and Wales have fluctuated somewhat over the last ten years<sup>17</sup>; Figure 1).



**Figure 1:** Annual totals of VTEC (*E. coli* O157:H7) infections in England and Wales.<sup>2</sup>

### Sources of infection

*E. coli* O157:H7 infections have been associated with a variety of sources and routes.<sup>11</sup> Apart from animal-to-person and person-to-person transmission, consumption of *E. coli* O157:H7-contaminated food, particularly in public places such as day care centres, is an important mode of transmission that has attracted much attention in recent years.<sup>11,18-20</sup> To date, research has investigated survival of the pathogen in a wide range of foods, including meat and meat products<sup>21,22</sup>, dairy products<sup>23</sup>, lettuce<sup>24</sup>, apples<sup>25</sup>, tomatoes<sup>26</sup>, chocolate and other confectionery<sup>27</sup>, and drinking water.<sup>28</sup>

Farm livestock, particularly ruminants like cattle, sheep, and goats, are regarded as the primary reservoirs for VTEC.<sup>29</sup> Numerous studies have investigated *E. coli* O157:H7 prevalence, transmission, survival and control in cattle and beef.<sup>11,22</sup> Among others, the review by Rhoades *et al.* (2009) discussed factors that influence the prevalence of three important pathogens<sup>22</sup>, VTEC *E. coli*, *Salmonella enterica*, and *Listeria monocytogenes* in the whole process of meat production. It is estimated

that the most severe cases of food-borne disease have been reported to be attributable to various foodstuffs containing beef. For instance, Adak *et al.* (2005) indicated that in England and Wales, 7% of the 1.7 million cases of food-borne disease in the period 1996-2000, including 67 deaths, were associated with beef.<sup>30</sup> In the Netherlands, undercooked ground beef and raw milk have most often been implicated in food-borne infections.<sup>29</sup> Different countries may present different situations of food-borne disease, depending on factors such as the pathogen load in the beef products consumed and the cooking and consumption habits of the country concerned.<sup>22</sup> Products such as lightly-cooked burgers may be eaten more frequently in the USA, while people in France and the Netherlands consume more steak tartare than people in the UK and Greece.

### Meat

*E. coli* O157:H7 exists as a normal coloniser of the gastrointestinal tract of cattle.<sup>29,31</sup> A number of studies have focussed on the prevalence of *E. coli* in the meat chain starting from the farm, the slaughterhouse, to the final, ready-to-eat products.<sup>22,31</sup> The spread of *E. coli* O157:H7 has been identified in farm housing and faeces<sup>32</sup> and the pathogen is known to survive for considerable periods in faeces and slurry.<sup>33</sup> This bacterium may readily leach from sheep and cattle faeces during rainstorm events thus leading to further infections.<sup>34</sup> Pigs and poultry can also be a source of O157 VTEC strain. Heuvelink *et al.* (1999) found that *E. coli* O157:H7 were isolated from 1.4% of 145 pigs and from 1.3% of 459 pooled faecal samples from turkey flocks but was negative in faecal samples from chicken flocks.<sup>35</sup> In a similar study, Kijima-Tanaka *et al.* (2005) isolated shiga toxin-producing *Escherichia coli* (STEC) from 23% of 62 bovine faecal samples and 14% of 25 swine samples and again there was no isolation from chicken samples.<sup>36</sup>

A Korean study by Jo *et al.* (2004) reported a higher prevalence of *E. coli* O157:H7 in cattle than in pigs (8.4% versus 0.3%) and none in chicken.<sup>37</sup> At slaughter, transmission of *E. coli* from faecal material and hides to carcasses varies from 4.5% to 56% and from 1.1% to 43.4% respectively, which poses a great threat for the contamination of raw meat with this pathogen.<sup>31</sup> In addition, contamination may occur during the dressing, skinning and evisceration phases.<sup>31</sup> Reinstein *et al.* (2009) examined the prevalence of *E. coli* O157:H7 in organically and conventionally raised beef cattle at slaughter and found 14.8% and 14.2% positives, respectively.<sup>38</sup>

An Irish study recovered *E. coli* O157 from 2.4% of beef trimmings samples, 3.0% of beef carcasses and 3.0% of head meat samples.<sup>39</sup> The probability of *E. coli* O157:H7 spreading during the mincing process may be highest in the meat chain.<sup>41</sup> One carcass contaminated with *E. coli* O157:H7 may quickly spread the bacterium across the whole batch of minced meat from uninfected cows. Cagney *et al.* (2004) detected *E. coli* O157:H7 in 2.8% of minced beef and beef burgers, both frozen and fresh<sup>41</sup>, in the Republic of Ireland. Magwira *et al.* (2005) investigated 400 meat samples (134 meat cubes, 133 minced meats,



133 fresh sausages) collected from 15 supermarkets and butcheries in Botswana and found prevalence rates of *E. coli* O157:H7 were 5.2 % in meat cube samples, 3.8 % in minced meat samples, and 2.3 % in fresh sausages.<sup>42</sup> In South Africa, the prevalence of *E. coli* O157:H7 was identified on selected meat and meat products (45 samples each of biltong, cold meat, mincemeat, and polony).<sup>43</sup> Strains of *E. coli* O157:H7 were isolated by enrichment culture and confirmed by polymerase chain reaction (PCR). Also investigated were the arteriogram profiles of the *E. coli* O157:H7 isolates. Five (2.8%) out of 180 meat and meat products examined were positive for *E. coli* O157:H7. A parallel study in Switzerland<sup>44</sup> was conducted on minced meat (beef and pork) samples to test for the presence of STEC. STEC was isolated from 2.3% minced beef samples and 1% minced pork samples.

#### **Dairy products**

Outbreaks of *E. coli* O157:H7 illness have been found to be linked with consumption of raw milk and cheeses made from unpasteurized milk.<sup>45,46</sup> In 1999, more than 11% of the total number of *E. coli* O157:H7 infections in England and Wales were due to unpasteurized milk and dairy product (CDSC, 2000, cited in<sup>46</sup>); most probably due to faecal contamination during milking (Hussein and Sakuma, 2005). Conedera *et al.* (2004) noted that although the prevalence of VTEC O157 in raw milk and cheese is low, the organism appears to be able to survive the various stages of the cheese-making process. They found that the heat treatment of milk at the beginning stages of cheese production is usually not sufficient to kill the contaminated vegetative bacteria which can later survive the manufacturing and curing procedures. For example, *E. coli* O157:H7 was found to survive during the manufacturing process of soft Hispanic-type cheese (Kasrazadeh and Genigeorgis, 1995). *E. coli* O157:H7 is characterized by its ability to survive in acidic environments (e.g. in cheddar cheese after a curing period of more than two months; Reitsma and Henning, 1996). Furthermore, fermented dairy products made from raw milk contaminated with *E. coli* O157 can pose a risk to human health.<sup>46</sup> Marek *et al.* (2004) examined the survival of *E. coli* O157:H7 in pasteurized and unpasteurized Cheddar cheese whey.<sup>47</sup> Five strains of *E. coli* O157:H7 were used for the study and were inoculated into 100 ml of fresh, pasteurized or unpasteurized Cheddar cheese whey at 10<sup>5</sup> or 10<sup>2</sup> CFU ml<sup>-1</sup>, and stored at varying temperatures. Results showed that survival of *E. coli* O157:H7 was significantly higher in the pasteurized whey compared to that in the unpasteurized samples at all storage temperatures. Stringent sanitary practices should therefore be undertaken, particularly during the storage and handling of whey and use of pasteurized milk for cheese manufacture.

#### **Vegetables and fruits**

In the past ten years, an increased number of *E. coli* O157:H7-related outbreaks have been associated with fresh produce such as lettuce, cantaloupe, and alfalfa sprouts.<sup>48-50</sup> This growing tendency could be due to increased consumptions of potentially risky fresh-cut pre-

packaged products.<sup>48</sup> Four separate outbreaks of food-borne *E. coli* O157 infections were recorded in USA in 2006.<sup>48</sup> Common vehicles of the disease noted were fruits and vegetables such as green-based salads, potatoes, lettuce, unspecified fruits, and sprouts.<sup>48</sup> Among the reported outbreaks, lettuce was the single most frequently mentioned produce.<sup>51,52</sup> Ackers *et al.* (1998) found 70% of patients in 40 Montana residents were infected with *E. coli* O157:H7 due to the consumption of purchased leaf lettuce.<sup>51</sup> In addition, Eribo and Ashenafi (2003) demonstrated that *E. coli* O157:H7 could be found in tomato and processed tomato products as well as products containing vinegar.<sup>26</sup> *E. coli* O157:H7 showed the ability to grow during germination a sprouting of alfalfa<sup>53</sup> and in acidic foods such as fermented Spanish-style table olives.<sup>54</sup>

#### **Resilience of *E. coli* O15:H7 to environmental conditions**

Environmental conditions such as temperature, pH value, water activity, and sodium chloride have important implications in the survival and growth rates of *E. coli* O157:H7 in foodstuffs. The bacterium is known to have a typical resistance to heat.<sup>55</sup> It can proliferate at a temperature range of 8-44.5°C, with the optimal temperature for growth at 37°C.<sup>56</sup> Cooking beef thoroughly to 71°C is effective in eliminating the organism<sup>57</sup>, although slow cooking of meats may not eradicate the organism as well as rapid heating.<sup>55,56</sup> Regardless of pH and water activity, survival of *E. coli* was found to be better at 5°C than at 20 or 30°C in tryptic soy broth (TSB).<sup>58</sup> Moreover, *E. coli* was found to survive but not grow during fermentation, drying, or subsequent storage at 4°C for 2 months.<sup>59</sup> The heat-resistant property of *E. coli* O157:H7 is relative as it can be influenced by many other environmental factors, including growth phase, the amount of heat applied, the rate of heating and the water activity.<sup>55</sup>

For instance, at 30°C, inhibition of growth of *E. coli* O157:H7 in TSB was enhanced by reduction of the water activity<sup>58</sup> as well as increase of sodium chloride concentration.<sup>60</sup> Much evidence has shown that pH value plays a primary role in the growth rates of *E. coli* O157:H7. For instance, growth rates are similar at moderate pH values (pH 5.5-7.5), but decrease significantly at lower pH values.<sup>56</sup> Yet, Benjamin and Datta (1995) found the organism to be acid tolerant under the optimal temperature (37°C), surviving at pH 2.5 for up to 7 h.<sup>61</sup> The pathogen is capable of acid-adaption and adapted cells have shown increased survival in shredded dry salami and apple cider.<sup>62</sup> *E. coli* O157:H7 has been reported to survive for months in acidic foods, such as fermented sausages<sup>63</sup> and apple cider and apple juice<sup>25</sup>; even though products such as fermented sausage may also lead to water stress in bacteria. The resilience of the organism to a combination of factors such as temperature, pH, water activity and sodium chloride can all contribute to the survival and growth of *E. coli* O157:H7 in foodstuffs. Its ability to withstand low pH environments is also of course crucial during passage through the gastro-intestinal tract of livestock and humans.

*E. coli* O157 can survive and grow in both aerobic and





anaerobic conditions as well as modified atmospheres used for food packaging.<sup>64</sup> As a facultative anaerobe, the heat resistance of this pathogen can vary between anaerobic and aerobic environments. For instance, it has been documented that there was little influence on the capability of *E. coli* O157:H7 under anaerobic conditions, but when aerobically-situated, the pathogen showed reduced heat-resistance.<sup>64</sup> Consequently, this has important implications in food packing. Therefore, there may be increased risk of *E. coli* O157:H7 surviving during heating treatments of foodstuffs that are packed under vacuum or reduced oxygen atmospheres.<sup>65</sup>

### Control of *E. coli* O157:H7

The increase in number of food-borne pathogenic infections has generated considerable efforts in the control of organism such as *E. coli* O157 in food. Many preventative measures have been introduced and targeted at all stages of the food chain, from the farm, to the slaughterhouse, and to the preparation of food at home.<sup>66</sup>

Although total elimination of *E. coli* O157:H7 carriage in livestock appears unlikely, pathogen transmission can be reduced through a number of farm management practices, such as to forbidding farmers from applying slurry and animal manure to vegetables and fruit plants.<sup>32</sup> Good hygiene practices such as careful preparation and cooking of food and interventions such as pasteurization, organic acid washes, and stream vacuuming, as well as the use of antimicrobial solutions (e.g. dilute lactic acid, trisodium phosphate and chlorine) can be effective means to eliminate *E. coli* O157:H7 from food.<sup>22,66,67</sup> For instance, to prevent minced meat from contamination with *E. coli* O157:H7 during the mincing process, cooking at a high temperature can destroy *E. coli* O157:H7 cells.<sup>43</sup> To prevent contamination of apple cider, it is suggested to wash and brush apples and preserve the cider with sodium benzoate (Zhao *et al.*, 1993; cited in Chapman, 1995) or aqueous commercial cleaner.<sup>68,69</sup> To reduce the number of VETC on salad vegetables, storing salad vegetables at 4°C can be an effective means.<sup>70</sup>

In recent years, advanced technologies have also been explored in the produce industry to reduce *E. coli* O157:H7 and other pathogens as well as to maintain the sensory quality of the produce itself.<sup>71</sup> In 2007, Muthukumarasamy and Holley investigated the effect of probiotic incorporation in dry fermented sausages before and after they were micro-encapsulated on the viability of *E. coli* O157:H7. The researchers found that there is a reduction in the viability of *E. coli* O157:H7. On the other hand, they reported that micro-encapsulation increased survival of probiotic strains, maintain sensory properties but reduced their inhibitory action against *E. coli* O157:H7. One study by Selma *et al.* (2008) showed the combined application of gaseous ozone and hot water could effectively control microbial growth in cantaloupe melon as well as maintain its initial sensory quality such as aroma and texture. However, this study failed to point out specific action of ozone in inactivating *E. coli* O157:H7. Mahmoud (2010) explicitly demonstrated the efficacy of

X-ray on inoculated *E. coli* O157:H7 (also including *L. monocytogenes*, *S. enterica* and *S. flexneri*) on shredded iceberg lettuce. By treating iceberg lettuce with 1.0 and 2.0 KGy X-ray, the study detected significant reductions of *E. coli* O157:H7 population in both conditions. This approach also showed its promising application because the sensory quality (i.e., visual colour) of leaves was not adversely affected during subsequent storage. Recently, the development of multistrain probiotic dairy products with good technological properties, has gained increased interest as protective cultures against infections.<sup>71</sup>

Although controlling *E. coli* O157:H7 in food through thermal treatment, chemical destruction and preventative interventions have showed some due efficacy, some studies also report negative findings. For instance, organic acids such as lactic acid and citric acid were reported ineffective in controlling *E. coli* O157:H7 in beef burgers, even when combined with freezing at -20°C for 2 hours.<sup>72</sup> Another study on traditional Iranian barbecued chicken (TIBC) reported that although essential oils of oregano and nutmeg showed effectiveness in inhibiting the growth of *E. coli* O157:H7 in a broth culture system, they reported no inhibitory effect against this pathogen in ready-to-cook TIBC, suggesting that *in vitro* investigation may not necessarily be applicable to food conditions.<sup>73</sup> Although the importance of temperature control and protective packaging has been emphasized in reducing pathogen growth on raw meat, inoculated *E. coli* O157:H7 strain NCTC 12900 could still increase when lamb chops were kept at 4 °C for 12 days.<sup>74</sup>

## CONCLUSION

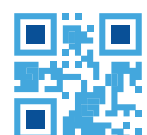
*Escherichia coli* O157 is an important food-borne bacterial pathogen closely associated with many severe human illnesses such as haemolytic uremic syndrome. Many of the intervention measures described are still effectively at experimental stage and are unlikely to be widely implemented in the foreseeable future due to a lack of commercial viability, geographical differences in the regulatory framework, or a lack of acceptance by consumers. Elevated public concerns about the adverse consequences of chemically synthesized preservatives used in food industry have diverted research to the application of natural antimicrobials to inhibit *E. coli* O157:H7 growth and activity.

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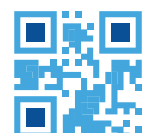
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