

EPI-Net Perspective

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Escherichia coli

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Introduction

Over the past year it seems the bacteria *E. coli* is everywhere. You have heard about them in news reports, it seems every day somebody is getting sick from eating undercooked hamburger meat, contaminated food or from swimming in contaminated water. But do all *E. coli* cause disease? The answer to this question is NO; in fact most *E. coli* bacteria are harmless or beneficial. For example, *E. coli* in the large intestine of humans is beneficial since it manufactures vitamin B12 while you digest food (2). So why the concern with this bacterium? The general answer is simple: there are two broad categories of *E. coli*, those that cause no harm but are used to indicate other possible problems and those that are capable of causing a disease.

Bacteria are small (1 µm in length) unicellular organisms that can live independently of a host. For example, because of their small size, a gram of soil can often contain over a billion bacteria. They have colonized a variety of environments ranging from the gut of a termite and most humans to Lake Michigan and beyond.

In this issue of EPI-Net Perspective we will discuss *E. coli*

What is *Escherichia coli*?

Escherichia coli, commonly abbreviated as *E. coli* is one of many bacteria present in the intestines of mammals. It was discovered in 1885 by Dr. Theodor Escherich, a German bacteriologist. *E. coli* is a gram negative, facultative anaerobe rod that belongs to the large family of bacteria called *Enterobacteriaceae*. This family includes: *Salmonella*, *Serratia*, *Shigella*, *Citrobacter*, and others. As a member of the intestinal flora, it is part of the digestive process and is excreted in feces. Shortly after an animal is born, its intestinal tract is colonized by bacteria.

Because it is excreted in high numbers in feces it is often used as an **indicator of fecal contamination**. The number of *E. coli* in feces averages from 10⁶ to 10⁷ per gram (wet weight) (11).

Historically, *E. coli* is included as a member of a group called fecal coliforms. There are two classifications of coliforms. General coliforms are defined as facultative anaerobe, gram-negative, non-spore forming, rod shaped bacteria that can ferment lactose



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to gas at 35°C (1). Fecal coliforms are all of the above but are also thermotolerant in that they can ferment lactose to gas at 44.5°C and includes the bacterium *E. coli*, *K. pneumoniae* and *Citrobacter*.

The presence of fecal coliforms has been used to suggest the occurrence of fecal material in water which in turns might indicate the presence of pathogens as they are often transmitted with fecal material from an infected host. More recently, the regulatory community has been moving away from the general term fecal coliform and is using the presence of *E. coli* as an indicator of fecal contamination.

Animals Manure



Food



Contaminated Water



Drinking Water



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E. coli as an indicator of fecal pollution

Characteristics of a good indicator: It should be non-pathogenic, rapidly detected and easily enumerated, should be associated with the presence of pathogenic organisms and have similar survival characteristics; it should not replicate in the environment and should be in greater numbers than the pathogens.

E. coli are now used as indicator of fecal pollution and to predict the presence of pathogens. If the source of fecal contamination (and hence the pathogen) could be appropriately identified, adequate implementation of best management practices could be allocated to reduce the transfer (⁹). While the direct detection of pathogens would give the best evidence of contamination, it is not always possible to detect pathogens as they are dilute, which makes detection more difficult since methodology is not sensitive enough to find the disease agent. Because of this, scientists have relied on the use of indicators that may predict the presence of pathogens in the environment.

What is in Fecal Material?

Fecal material is 75% water and 25% solid material. This solid material is composed of: dead and live bacteria, undigested food, fats, salts and waste material from blood. Some examples of normal intestinal flora bacteria are: *Bacteroides fragilis*, *Lactobacillus*, *Clostridium perfringens*, *Staphylococcus aureus*, *Enterococcus faecalis* and *Escherichia coli* among others.

E. coli has been selected as an indicator of fecal pollution because both its occurrence in human and animal feces. Also there are a myriad of affordable, fast, sensitive, specific and easy methodology to identify *E. coli*, making methodology accessible in to all⁽¹⁰⁾. Some of the methodology includes PCR (Polymerase Chain Reaction), specific growth media such as Colilert® or Colisure® and EMB (Eosin Methylene Blue Agar), and biochemical tests such as Citrate among others. Once the presence of *E. coli* has been confirmed, another cascade of methods have been developed in order to determine its origin, some examples are: ARA (Antibiotic Resistance Analysis), Rep-PCR (Repetitive Extragenic Palindromic element PCR) and Ribotyping.

For example: documented data for the pathogen *Salmonella*, shows levels to be increased at high levels of total coliforms ($\geq 10^4$ CFU/100 ml), and to occur at low levels when total and fecal coliform levels are low (25 CFU/100 ml of total coliforms and 13 CFU/100 ml of fecal coliforms) (⁷). Therefore it is assumed that if a waste material contains *Salmonella*, high levels of fecal coliforms are likely to be present.



E. coli as a pathogen

E. coli are usually harmless but some strains of *E. coli* are pathogenic. There are 9 classes of disease causing *E. coli*:

1. **Enterotoxigenic *E. coli*** (ETEC) - this strain is usually associated with diarrhea among third world children and traveler's diarrhea in adults, it is usually transmitted by ingestion of contaminated food or water. ⁽⁶⁾

2. **Enteroinvasive *E. coli*** (EIEC) - this strain can invade the colonic epithelium and causes diarrhea. It is transmitted by ingestion of contaminated food and water as well as contact with contaminated individuals ⁽⁶⁾.

3. **Enterohemorrhagic *E. coli*** (EHEC) - originally believed to be an evolved strain of enterotoxigenic *E. coli*. It is a food borne pathogen ⁽⁸⁾ and has been usually associated with the consumption of undercooked hamburger. *E. coli* O157:H7 belongs to this group ⁽⁶⁾. Shiga toxins - is the virulence factor in EHEC, (Stx), and the factor responsible for the complications in infected people. *E. coli* strains carrying the Stx toxin are usually found in feces of animals like cattle, sheep cats, dogs, chickens, pigs, goats and gulls. Shiga toxins are part of a large family of plant and bacterial toxins that cause cell death by the inhibition of protein synthesis. *E. coli* strain O157:H7 is the one related with most infections but other shiga strains have been reported, i.e., O26:H1, O103:H2, O111: NM and O113:H21.

4. **Enteropathogenic *E. coli*** (EPEC) - causative agent in children's diarrhea ⁽⁸⁾, usually infecting children under 2 years of age ⁽⁶⁾. Since this type of strain can be isolated from both healthy kids (older than 2 years) and adults; humans are a major reservoir. Infection can be cause by the fecal-oral route through contaminated food or hands ⁽⁶⁾.

5. **Enteroaggregative *E. coli*** (EAaggEC) - It is associated with diarrhea of infants in develop and undeveloped countries. First recognized by a clumping due to a distinctive adherence to Hep-2 cells ⁽³⁾.

6. **Uropathogenic *E. coli*** (UPEC) - is the major cause of urinary track infections (UTI). It is usually the result of contamination from feces.

7. **Neonatal Meningitis *E. coli*** (NMEC) - is a result of an invasion of *E. coli* to the meninges through the blood stream. About 80% of NMEC *E. coli* strains contain the K1 capsular polysaccharide virulence factor ⁽¹⁴⁾.

8. **Verotoxigenic (VTEC)** - verotoxin producing *E. coli*. Verotoxin is a type of shiga toxin producing bacteria (similar to those present on ETEC). They are known to cause oedema disease (ED) in weaned pigs and diarrhea in humans. Contaminated food (cattle) is usually the vehicle for infection with this type of pathogenic *E. coli* strain ⁽⁵⁾.

9. **Necrotoxigenic *E. coli*** (NTEC) - is a recently discovered pathogenic *E. coli* who has the capacity of producing cytotoxi necrotising factor (CNF) ⁽¹²⁾. CNF is a toxin that induces necrosis (cell death). There are two types: NTEC 1 and NTEC 2. NTEC 1 has been related to diarrhea, septicemia and urinary track infections in humans, cats, dogs and pigs, whereas NTEC 2 affects mostly ruminants causing diarrhea and septicemia ⁽⁴⁾.

Summary: *E. coli* are both friend and foe. They contribute to human and animal health by converting food to the vitamins we need. Because they occur in high numbers, they can be used to indicate the recent introduction of fecal material to water. However, some data indicates that *E. coli* can grow and function in the environment so this brings into question the timing of the introduction ⁽¹³⁾. On the other hand, certain types are capable of causing significant diseases and even death. To be on the safe side we must manage our natural resources to eliminate any *E. coli* introductions.



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Glossary of Terminology

Facultative anaerobe – organisms that can grow with or without oxygen but grow better in its presence.

Gram negative bacteria – gram stain is a procedure that distinguished bacteria based on their ability to retain crystal violet when decolorizing with an organic solvent such as ethanol. The gram negative type loses the crystal violet, staining the cells pink. Gram negative bacteria have their cell wall composed of a peptidoglycan layer surrounded by lipopolysaccharide.

Thermotolerant bacteria – organisms that are capable to grow at higher than optimal temperatures without shutting down their metabolism.