Escherichia coli Strains in Patients with Inflammatory Bowel Diseases: A Review

Hadba Al-Amrah*, Hanan Alotaibi and Nemat Sadiq

Department of Biological Sciences, Faculty of Science, King Abdulaziz University, Jeddah-21589, Saudi Arabia.

http://dx.doi.org/10.13005/bbra/2966

(Received: 30 December 2021; accepted: 08 March 2022)

Gastrointestinal tract conditions, including inflammatory bowel diseases (IBDs) such as ulcerative colitis (UC) and Crohn’s disease, have been linked to adhesive invasive Escherichia coli (AIEC) pathotypes, with comparable pathogenic properties, although the incidence of AIEC with UC and CD is generally undetermined. While a significant advance has been made in understanding the pathogenic processes of AIEC since it was first characterized a decade ago, the molecular basis that determines the phenotypic features of AIEC pathotypes is still unknown. This article reviews studies that examine the prevalence of E. coli in patients with IBD and discusses its pathophysiological role.

Keywords: Adherent Invasive Escherichia Coli; Crohn’s Disease; Epidemiology; Inflammatory Bowel Disease; Pathogenesis.

Inflammatory bowel disease (IBDs), most predominantly Crohn’s disease (CD) and ulcerative colitis (UC), subtypes, are intestinal conditions that involve persistent inflammation of the gastrointestinal tract. IBD is a severe chronic inflammatory illness of the intestine affecting more than 0.3% of people, with UC and CD being the most common. Internationally, IBD is more common in wealthy countries although is becoming increasingly prevalent in developing countries. Although the cause of IBD is not fully determined, a growing body of research suggests morbidity is strongly linked to hereditary susceptibility. Additional variables, such as nutrition, tissue damage linked with immune system disturbances, and aberrant gut microbiota, may be implicated, as evidenced by mouse models of IBD. It is important to note that the genetic and microbiota-related origin of IBD may be connected. Recently, scientists may have made a fascinating discovery that points to a probable cause of CD: individuals with CARD15/NOD gene mutations, which rely on lower nuclear factor kappa B activation (NF-κB), proinflammatory cytokine production, and defensin secretion, are prone to CD development.

Inflammatory Bowel Disease and the Intestinal Bacteriome

IBDs have confounded immunologists and gastroenterologists since their first description between 75 and 100 years ago. Currently, novel investigative approaches are rapidly leading to improved knowledge of key pathophysiologic mechanisms connected to these disorders, paving the way for effective therapies. Patients with UC report abdominal discomfort, diarrhea, weight loss, rectal bleeding, fever, and exhaustion as gastrointestinal and systemic symptoms. Patients
with CD patients can evolve intestinal strictures and fistulae among parts of the intestinal and between the colon, skin, and other organs. The symptoms of UC are virtually identical to those of CD excepting no formation of fistula with UC. Both UC and CD are usually chronic and relapsing11.

In humans, the gut microbiota composition includes about 1000 bacteria species, five Archaea genera, 66 fungus genera, and an ill-defined number of viruses, predominantly bacteriophages. These are essential for the immune system and other physiological systems which cannot function without this intricate ecosystem. However, the gut microbiota does not initiate intestinal inflammation independently, rather, abnormalities in the microbiota (i.e., dysbiosis) or the presence of commensal bacteria, with higher virulence in IBD patients, may trigger an overactive antimicrobial immune response12. CD involves an extreme interleukin (IL-12/IL-23) and interferon-gamma/interleukin-17 (IFN-ã/IL-17) production in the small bowel and full-thickness intestinal wall inflammation and discontinuous colon ulceration, which frequently includes granulomas. By contrast, UC is linked to excessive IL-13 production and principally influences the colon, with a continued mucosa inflammation involving the rectum and extending proximally11 (Fig.1).

While strongly correlative in terms of presentation and symptoms, both CD and UC have distinct clinical characteristics. There are various serums that differentiate the primary types of IBD and reflect their activity or treatment. Furthermore, an evaluation of anti-microbial antibodies and T cell response to commensal gut bacteriome prevalent in healthy people found significant differences with CD patients13. These are important distinctions between CD and UC that suggest that angiogenesis and inflammation regulation may differ. Thus, IBDs occur in immunocompromised patients (such as chronic granulomatous illness and variable immunodeficiency) with hereditary conditions (like Hermansky-Pudlak syndrome), meaning various immune components have a role in IBD susceptibility. Many researchers examining the pathogenesis of IBD believe that it is caused by an interaction between the gut’s bacterial microflora and the mucosal immune system14. Thus, the bacterial microflora plays an essential role in the etiology of IBDs; if the microflora is quantitatively and qualitatively normal, the disease defect is found in the mucosal immune system15. The normal state of immunologic tolerance to microbial antigens in the gut is disrupted in IBD cases, either by the presence of a defective mucosal effector T cell population that overreacts to usual bacterial antigens or by the presence of a defective mucosal T-regulatory cell population that under-reacts to normal microorganism antigens16. An alternative cause is that the gut microflora has a fundamental abnormality in the abundance or kind of microbiota that make up the community, or the degree to which the organisms interact with the mucosal immune system is abnormal. Despite the fact that numerous microorganisms have been explored as causative factors in the aetiopathogenesis of IBD, the disease can lead to a loss of tolerance since the microbiota is able to drive a normal immune system to respond excessively to microbial antigens. Mycobacterium paratuberculosis, Listeria monocytogenes, Chlamydia pneumonia, Escherichia coli (E. coli), and other bacteria are among these pathogens. The dynamic equilibrium between intestinal bacteria, particularly commensal flora, and the host defense systems at the intestinal mucosa, as well as their role at the outset and connection with intestinal inflammation, are currently receiving greater attention16. Also, changes in the gut bacterial flora caused by environmental, and specifically dietary, factors are thought to have a significant role in IBD etiology17.

**Differences In Gut Microbiota In IBD**

IBD is an autoimmune condition that necessitates the presence of commensal bacteria in the gut. Many studies have propelled the idea that IBD infections are caused by an overactive immune response to a normal member of the gut microbiota18. Duchmann et al. found cells derived from gut IBD tissue when cultured with sonicates of autologous or heterologous intestinal microbes displayed stimulation; by contrast, the cells from the control respond to sonicates of heterologous microbiota only19. The most widely sketched pathogenesis of IBD is that the sickness is largely a result of the abundance of specific microbiota which triggers a pathological immunological response in the mucosal immune system20. There are two bodies of evidence(3) that support the autoimmune response theory. The first suggests that IBD is linked to pathogenic
organisms that cause a low-grade infection of the mucous and as a result elicit an inflammatory response. The second shows that patients with IBD have a defective epithelial barrier that allows nonpathogenic organisms to increase close to parts of the mucosal immune system, provoking inflammatory response\(^1\). To assess the first body of evidence, we can examine the findings of recent studies on microbiota linked with the mucosa (rather than the stool microbiota), which are more likely to give rise to infection. A number of studies have found that biopsy tissue from patients with IBD had greater levels of mucosa-linked bacteria in the mucus layer and on the epithelial tissues than tissue of healthy people\(^1\). In IBD research, many studies have found an increased abundance of mucosa-associated microbiota\(^2\)-\(^2\)\(^3\). In one case, a pathogen-like invasive \textit{E. coli} was found in the mucosa of 20\% and 40\% of ileal mucosa samples from patients with CD compared to 6\% found in the mucosa of samples from healthy individuals. Also, invasive bacteria were found in roughly 4\% of colonic samples from control patients and CD patients compared to 12\% of patients with UC samples\(^2\)\(^4\). A study by Martin et al., however, has cast doubt on the significance of such findings, reporting that the majority of patients with CD and a significant number of healthy people, have substantially higher rates of mucosa-adherent microorganisms (80\% in CD patients and 40\% in healthy individuals)\(^2\)\(^4\). Dietary types and environmental upset that would ordinarily affect species structure and short chain fatty acid (SCFA) levels frequently have a significant effect on the gut microbiota. IBD is marked by long-term alterations in the gut microflora, which are linked to inflammation in the intestine. A substantial drop of butyrate-producing obligate anaerobes from the Firmicutes phylum, the most abundant of which is \textit{Faecalibacterium prausnitzii}, and is a typical biomarker of IBD\(^2\)\(^5\), \(^2\)\(^6\). Since dysbiosis of obligate and facultative anaerobic bacteria characterize IBD imbalance, it has been argued that oxygen and reactive oxygen species have an essential role in its pathogenesis. Indeed, according to the ‘oxygen hypothesis’, prolonged intestinal inflammation causes an increase in the liberation of oxygen-carrying hemoglobin and reactive oxygen species within the lumen, resulting in a milieu that promotes facultative anaerobic microorganisms. Increased inflammation results from the decrease of obligate anaerobes like \textit{F. Prausnitzii} that release

![Diagram](image_url)

**Fig. 1.** Comparison of the mucosal immune response in healthy individuals and CD patients with faulty nucleotide-binding oligomerization domain containing 2 (NOD2) function, taken from (10)
anti-inflammatory chemicals, causing a feedback loop that exacerbates the disease process\textsuperscript{16, 27} (Fig. 1).

The toll-like receptor 2 (TLR2) on the surface of dendritic cells in the gut lamina propria detects peptidoglycan (PGN) produced on commensal bacteria walls (left inset, Fig. 1).ensed by TLR2, and as a result, there is NF-50B downstream activation, which is a critical transcription factor essential for the recognition of cells that produce IFN-\(\alpha\) and IL-17, the pro-inflammatory cytokines that are thought to cause CD. PGN, by contrast, is broken down in endosomes and so serves as a source of muramyl dipeptide (MDP), a molecule that nucleotide-binding oligomerization domain containing 2 (NOD2) detects and activates. As a result of this activation, PGN-mediated NF-50B activation is inhibited, resulting in down-regulation of TLR-induced cytokine production. Since NOD2 modulation is disrupted in individuals with CD that have NOD2 mutations (right inset, Fig.1), the innate immune ‘thermostat’ of the gut is set at a greater level of proinflammatory cytokine production. Inflammation and disease stem from this and T cell response to mucosal antigens.

Increased bacterial abundance in the terminal ileum’s crypts results in greater activation of a mucosal immune system already functioning at a higher grade\textsuperscript{10}.

\textbf{E. Coli Pathogenicity}

Within a few hours of birth, \textit{E. coli} colonizes the gut system of newborns. \textit{E. coli} and its mammalian host colon co-exist in harmony, known as mutualism. It is a strong rival in the gut microflora against anaerobes and facultative anaerobes; it is excluded in immunocompromised hosts or when the usual gut barriers crossed, although \textit{E. coli} seldom cause illness. Escherich, however, has claimed that specific \textit{E. coli} might be linked with disease, indicating which \textit{E. coli} are implicated in infections of the colon and urinary system. Through DNA horizontal transfer of transposons, bacteriophages, and plasmids, particular \textit{E. coli} have gained unique virulence factors. These improve the capacity of this bacteria to fit into new habitats, allowing them to cause a wider range of illnesses. There are six gastrointestinal tract pathogenic \textit{E. coli} (IPEC) bacteria linked to GI tract diseases in humans: enteroinvasive \textit{E. coli} (EIEC), enteropathogenic \textit{E. coli} (EPEC), enterotoxigenic \textit{E. coli} (ETEC),

\begin{figure}[h]
\centering
\includegraphics[width=\textwidth]{fig2.png}
\caption{Classification of \textit{E. coli} into six recognized diarrheagenic categories by virtue of distinctive features that influence eukaryotic cells, taken from (31)}
\end{figure}
Table 1. Studies on changes in gut E. coli quantity in IBD patients

<table>
<thead>
<tr>
<th>Ref.</th>
<th>Type of sample</th>
<th>Method</th>
<th>Abundance of E. coli in CD and UC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clapp et al., 2017</td>
<td>Mucosa</td>
<td>quantitative Polymerase Chain Reaction (qPCR)</td>
<td>Increase in E. coli abundance in CD patients</td>
</tr>
<tr>
<td>Lopez-Siles et al., 2014</td>
<td>Mucosa</td>
<td>qPCR</td>
<td>Increase in E. coli abundance in CD patients</td>
</tr>
<tr>
<td>Sha et al., 2013</td>
<td>Feces</td>
<td>qPCR</td>
<td>Increase in E. coli abundance in CD and UC patients</td>
</tr>
<tr>
<td>Reichert et al., 2010</td>
<td>Mucosa</td>
<td>qPCR</td>
<td>Increase in E. coli abundance in CD patients</td>
</tr>
<tr>
<td>Martinez-Medina et al., 2009</td>
<td>Biopsies and mucosa</td>
<td>qPCR</td>
<td>Increase in E. coli abundance in CD patients</td>
</tr>
<tr>
<td>Baumgart et al., 2007</td>
<td>Mucosa</td>
<td>FISH 16S rRNA clone</td>
<td>Increase of E. coli strains in CD patients</td>
</tr>
<tr>
<td>Mylonaki et al., 2005</td>
<td>Biopsies</td>
<td>FISH</td>
<td>Increase of AIEC in CD and UC patients</td>
</tr>
</tbody>
</table>

enteroaggregative E. coli (EAEC), diffusely adherent E. coli (DAEC), and enterohaemorrhagic E. coli (EHEC)(2). These pathogenic E. coli cause illness via affecting a vast extent of key host cell activities, such as protein synthesis, apoptosis, signal transduction, mitochondrial role, ion secretion, transcription, cytoskeletal role, and cell split. Moreover, regarding toxins and effectors of microorganisms that impact eukaryote processes, all these pathogens must express a variety of fitness and colonization factors that permit the microorganisms to cling to host cells (28). Several studies based on various techniques have found that E. coli is increased in patients with IBD\textsuperscript{16,29,30} (Table 1)

**Adhesive Characteristics of IBD-Linked E. Coli**

It has been found that gut colonization by E. coli is linked to bacteriome adherence by CD-linked E. coli to digestive tract mucosa (to the epithelial cells). The initial stage in the pathogenicity of many organisms with gastrointestinal infections is bacterial adherence to intestinal epithelial cells. The action of the bacteria to colonize the epithelial cells and combat mechanical sweep from the gut is achieved by adhesion. The broad conclusion across a number of studies is that IBD-related adhesion to E. coli can attach to several human cells. According to one study\textsuperscript{12}, sticky E. coli was identified in 62 % of CD patients and 68 % of UC patients, but only 6 % of healthy control\textsuperscript{12}. In another study, 86 % of E. coli isolated from patients with IBD were sticky, compared to 27 % of E. coli isolated from patients with infective diarrhea and healthy controls\textsuperscript{12}. Recently, according to Kotlowski et al.\textsuperscript{35}, E. coli with adhesion factors in CD and UC patient tissues were more abundant than the same in healthy controls. When E. coli bacteria was recovered from the ileum of patients with CD and also from healthy people, it was found that nearly 80 % of E. coli correlating with the ileal mucosa of patients with CD were adhesive compared to 30 % separated from healthy individuals\textsuperscript{16}. CD-linked E. coli attached to distinguish Caco-2 cells, preferably a mature gut cell sample\textsuperscript{16}. This is in line with the discovery that crypt epithelial cells related to immature cells are seldom implicated in patients with early lesions in connection with CD.

**Invasive Characteristics of IBD-Linked E. Coli**

The lesions that form in CD exist in Peyer’s patches, as seen in early entero-invasive
microorganisms, such as Shigella and Salmonella, which can induce GI tract lesions. This supports the notion of an invasive pathogen that initiates CD. Indeed, the aphthous ulcer, the necrosis of microfold cells (M-cells) of Peyer’s lymphoid follicles, is identified as the earliest lesion of CD. Invasive shigelloides, salmonellosis, yersinia enterotoxigenic, and colitis are primary associated virulence factors of the disease. Many studies have reported the existence of intramucosal E. coli in patients with IBD or mucosa-associated E. coli with invasive traits. Invasive microbiota exists in 29-36% of patients with CD, in 12-19% of UC patients, and 3-9% of healthy individuals. The invasive process of LF82 strains separated from a lesion of a CD patient has been extensively studied. LF82 strains efficiently pervade many human epithelial cells, including HEp-2 cells, HCT-8 cells, and the gut cell lines intestine-407 and Caco-2. The most invasive bacteria, Yersinia enterocolitica, Shigella Flexner, and Listeria monocytogenes, and EIEC, are actin microfilament, but not microtubule-dependent. The uptake of the invasive E. coli and LF82 separated from patients with CD is based on the role of host cell microtubules and actin microfilaments. A micropinocytosis-like method of entry was discovered in LF82-infected epithelial cells, distinguished by elongation of the membrane extensions that encircled the bacteria at the areas of contact among the epithelial cells and the entering bacteria. The LF82 strain survives and multiplies in the host cell cytoplasm after lysing the endocytic vacuole. The invasive mechanism of LF82 is unique in that it lacks any of the renowned genetic invasive determinants seen in entering invasive, Shigella strains enteropathogenic E. coli and enterotoxigenic E. coli. Also, Pseudomonas aeruginosa and Helicobacter pylori outer membrane vesicles are shown to construct proinflammatory responses, and TLR5, when it reacts with bacterial flagellin, can activate an innate immune reaction.

Replication of CD-Correlating E. coli in Microphages

Intracellular infections have evolved to combat phagocytosis and survived interior macrophages. The macrophage activation and engagement in persistent antigenic stimulation cells have been the center of the hunt for pathogenic organisms that may prompt CD. Invasive E. coli obtained from CD may live and multiply in a large vacuole of murine macrophages. CD-linked invasive E. coli behaves differently in macrophages than other invasive microbiota. While most members of invasive bacteria cause cell death in macrophages that are infected, macrophages infected with CD-linked invasive E. coli show no necrosis or apoptosis, even after 24 hours. Moreover, CD is related to invasive E. coli that are picked up within macrophages by phagosomes that develop without diverging from the conventional endocytic route and that engage with phagolysosomes. By contrast, many pathogens infiltrate autophagy or escape the normal endocytic process. Microbiota have evolved mechanisms where acidity is a critical sign for triggering the expression of malignancy genes and increase in the severe environment found within these compartments, including cathepsin D proteolytic activity and acid pH. Tumor necrosis factor (TNF) is released in substantial levels by macrophages infected with CD that are related to invasive E. coli. TNF is transcribed and translated from scratch following macrophage activation, indicating that macrophages are still active despite many intracellular bacteria. The persistent proliferation of internal bacteria within the phagosomes causes continuous activation and TNF release.

Pathogenic Properties of E. Coli

Isolated E. coli bacteria from IBD patients are clonally heterogeneous, belonging to multiple serotypes and sequence types. Though a close genetic hereditary link has been found in pediatric patients with IBD, the concept of IBD that is caused by a specific clone has been generally dismissed. In contrast, E. coli is identified in patients with IBD in combination with extraintestinal pathogenic E. Coli; typically B2 and D phylotypes. Extraintestinal pathogenic E. coli. Many studies show that B2 and D phylotypes colonize IBD patients more than healthy people, whereas some other studies suggest that IBD and healthy participants have similar phylogroup distributions. These differences in findings could be a product of the different types of samples studied, since it has been observed that transitory E. coli, the most abundant type discovered in stools, B1 and A phylotypes specifically, are commonly found in healthy people. By contrast, resident E. coli, which is the most...
common identified from biopsy, results mainly D and B2 phylotypes, which are generally associated with IBD. As a result, investigation depending on biopsies samples will tend to show B2 and D strain abundance, even in healthy people. Another factor that alters perceived abundance of phylotypes in IBD is illness severity^{46}. A higher fraction of B2 and D separate has been detected in inactive IBD patients, which has been linked to inflammatory level of tissue^{44, 47}. This indicates a change in abundance of *E. coli* toward separates that are more suited to inflamed tissue in IBD patients or are active in the inflammatory process^{48}. To the present authors’ knowledge, there have been no reports of phylotype distribution discrepancies among UC and CD patient *E. coli* strains that have distinct sets of virulence genes than pathogenic *E. coli* (ExPEC) strains. Nevertheless, intestine ExPEC is very rare or perhaps nonexistent^{49}. The presence of virulence factors in *E. coli* in healthy people is thought to be crucial for the effectiveness of colonization in the gut mucosa. Malignancy gene profiles are inextricably related to the evolutionary origin of the strain^{47}. Where B2 and D are predominant in patients with IBD, more commonly discovered virulence-linked genes indicative of ExPEC were found in IBD patients than in controls, depending on the abundance of phylogenetic groups, and without differences in other investigation types^{50}. A shift in the phylotype distribution would result in increased abundance of *E. coli* with colonization factors, facilitating fixation and persistence in IBD patients. However, it is unknown if the alteration occurs only in IBD patients or is a universal trend in industrialized nations. Although inspecific genetic characteristics separate *E. coli* from the UC or CD gut mucosa, certain virulence factors have been discovered that are distributed variably amongst these IBD types. For instance, diarrhea-linked hemolytic *E. coli*, also known as cell-detaching *E. coli* (CDEC), has been detected in about 24 % of UC *E. coli* patients, while detected in only 4.7 % of CD *E. coli* patients^{50}. Diarrhea-linked hemolytic *E. coli* usually carries pilus P, hemolysin, S-fimbria genes, and cytotoxic necrotizing factor 1. The uropathogenic-specific protein (USP) gene, which codes for the uropathogenic-specific protein discovered in UC patient *E. coli*, is more abundant than in CD patient *E. coli*^{50}. The *E. coli* containing iro gene, which codes for an iron-chelating siderophores receptor, was newly seen to be more very often separated from inflammatory and un inflamed mucosa-active patients with UC. Darfeuille-Michaud et al. found a novel pathotype of *E. coli* with different phenotypic pathogenic characteristics that were not linked with UC but associated with CD, and termed adherent invasive *E. coli* (AIEC)^{51}.

**CONCLUSION**

IBDs are becoming more common throughout the world, in developing and developed countries. Although the causes are undetermined, a burgeoning body of research has found a clear link between IBD morbidity primarily due to hereditary susceptibility, although in connection with environmental factors, and have shown key characteristics and biomarkers of the gut microbiota that characterize pathogenesis and susceptibility. Variables such as nutrition, tissue damage linked with immune system disturbances, and aberrant gut microbiota have been implicated. *E. coli* has been found to be an essential microorganism of a healthy gut but can have a key role in the development of IBD.

**ACKNOWLEDGMENTS**

We would like to extend our thanks to everyone that assisted in the English language revision and editing and proofreading of the manuscript.

**Conflict of interests**

The authors declare no conflicts of interest.

**Funding**

No funding source.

**REFERENCES**

3. Baumgart M, Dogan B, Rishniw M, Weitzman G,
Host Immune Response in Inflammatory Bowel Disease. *Front Immunol*. 2017;8:942-


2019;2019:9708494.


