Environmental Risk Factors for Inflammatory Bowel Disease

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Abstract: Inflammatory bowel disease (IBD) is characterized by chronic inflammation of the gastrointestinal tract and is associated with significant morbidity. The etiology of IBD has been extensively studied during the last several decades; however, causative factors in disease pathology are not yet fully understood. IBD is thought to result from the interaction between genetic and environmental factors that influence the normal intestinal commensal flora to trigger an inappropriate mucosal immune response. Although many IBD susceptibility genes have been discovered, similar advances in defining environmental risk factors have lagged. A number of environmental risk factors have been explored, including smoking, appendectomy, oral contraceptives, diet, breastfeeding, infections/vaccinations, antibiotics, and childhood hygiene. However, most of these factors have demonstrated inconsistent findings, thus making additional studies necessary to better understand the etiology of IBD.

Inflammatory bowel disease (IBD), which consists of Crohn's disease (CD) and ulcerative colitis (UC), is a complex genetic disorder that is influenced by environmental risk factors. The importance of genetic susceptibility has been established through genome-wide association scans, which have identified susceptibility genes (such as *NOD2* gene variants)[1,2] linking the pathogenesis of IBD to the dysregulation of the gastrointestinal immune system and the host microbiome.[3,4] Genetic predisposition, however, cannot be solely responsible for disease etiology. The lack of complete penetrance must be accounted for by additional factors in disease etiology.[5] Additionally, genetics cannot account for the rapid rise of IBD incidence in certain geographic regions.[6,7]

IBD has been primarily characterized as a disease of industrialized nations, with increased prevalence in the developed world. Since the 19th century, the incidence of IBD has increased steadily in North America and Europe until stabilizing in the middle and latter parts of the 20th century for UC (2–15/100,000 person-years) and CD (3–15/100,000 person-years), respectively. Although developing regions have traditionally reported lower prevalence of IBD, the incidence of IBD is rising in many of these nations (eg, India and China) as they have become industrialized.[8,9] Furthermore, migrant studies have demonstrated that individuals immigrating from regions with low prevalence to countries with higher prevalence are at an increased risk of developing IBD, particularly

among first-generation children.[5,6] Thus, environmental exposures are thought to contribute to the development of IBD.

Several theories of environmental causes of IBD have been postulated, and numerous environmental risk factors have been explored. In this paper, we will summarize the current knowledge of the association between the most commonly studied environmental exposures and IBD development.

Hygiene Hypothesis (Table 1)

Multiple theories have been proposed to explain the unknown environmental exposures that may interact with the immune system and result in an abnormal inflammatory response to intestinal microflora.[10] The most predominant theory is the hygiene hypothesis. This hypothesis proposes that the rising frequency of immunologic disorders can be attributed to a lack of childhood exposure to enteric pathogens.[5,11] Improved sanitation and hygiene, along with decreased exposure to enteric organisms during early childhood, may lead to a greater susceptibility to develop an inappropriate immunologic response upon exposure to new antigens (eg, a gastrointestinal infection) later in life.[12] Many factors have been examined as proxy markers of environmental exposures in early life, including *Helicobacter pylori* infection, family size, sibship, birth order, urban upbringing, and pet exposure.[10,13-15]

Helicobacter pylori

H. pylori is an infection often acquired early in childhood. Colonization has been correlated to sibship size, household crowding, and poor sanitary facilities.[16] IBD is more prevalent in developed nations where *H. pylori* infection is less common.[17] A meta-analysis of 23 studies concluded that *H. pylori* infection was negatively associated with both CD and UC.[17] Furthermore, colonization may protect against other immune conditions such as asthma.[17] *H. pylori* infection may protect against the development of IBD by increasing the expression of genes (eg, *FOXP3*) that are involved in T-regulatory cell function.[17] However, reduced colonization of *H. pylori* in IBD patients may be secondary to antibiotic and mesalamine use,[18] which could eradicate *H. pylori* infection in some IBD patients.

Family Size, Sibship, and Birth Order

Individuals raised with fewer siblings may have fewer opportunities to acquire the enteric infections during childhood that are necessary to program the immune system of the gut to respond appropriately to bowel infections later in life.[12,18] In one population-based study, CD patients were shown to live in smaller households and with fewer siblings[15]; however, similar results were not found for UC.[15] Other studies have demonstrated that IBD patients were raised with a greater number of older siblings than control patients[18] and that the number of older siblings conferred an incremental increased risk of developing UC.[19] Lower birth rank was associated with an increased risk of both CD and UC.[20]

Urban Environment

Children raised in urban societies tend to have a more "hygienic" upbringing than those living in rural locations (eg, on a farm). Differences in lifestyles and environmental exposures (eg, diet) in urban versus rural areas may contribute to the higher occurrence of IBD in urban areas.[21] Several observational studies have shown an increase in UC and CD incidence in more densely populated areas.[22-27] Numerous studies, however, have failed to find any association between urban exposure and IBD. A population-based case-control study,[28] as well as a later study conducted by Malekzadeh and associates,[29] failed to find a relationship between urban environment and either CD or UC. Further complicating the understanding of this relationship is a case-control study in the United Kingdom that found an inverse relationship between urban environment and CD but no relationship with UC.[16] These findings are supported by a study in France that used spatial analysis to show that CD was more common in periurban and rural areas.[30]

Other Childhood Factors

Other factors that support the hygiene hypothesis include a decreased risk of IBD associated with living on a farm, drinking unpasteurized milk, and housing density, though these findings were more commonly reported in CD compared to UC.[13-15] CD was more than 3-fold more common among those whose childhood homes had a hot-water tap and a separate bathroom.[12] Additionally, CD patients diagnosed in adulthood have been shown to be significantly less likely to have lived with pet cats before 5 years of age[15]; however, exposure to cats in early life was shown to be a risk factor in pediatric-onset CD.[14]

Specific Risk Factors for Inflammatory Bowel Disease (Table 1)

Numerous environmental risk factors of IBD have been explored, including smoking, oral contraceptive pills (OCPs), appendectomy, diet, breastfeeding, and nonsteroidal anti-inflammatory drugs (NSAIDs); however, none of these factors completely explain the environmental determinants of IBD.

Smoking

A paradoxical relationship has been consistently demonstrated between smoking and IBD. A meta-analysis

concluded that active smokers were less likely to develop UC compared to individuals who were never smokers or ex-smokers; in contrast, active smokers, followed by ex-smokers, were at an increased risk for acquiring CD.[31] A dose-response relationship between smoke exposure and IBD has been described.[32-34] The exact mechanisms by which smoking influences the development of IBD are unknown. Nicotinic acetylcholine receptors (nAChRs) are present in mucosal epithelial cells of the bowel.[35,36] The expression of nAChRs has also been found on T cells, indicating that nicotine may directly regulate T-cell function.[37] However, clinical trials of nicotine replacement in UC have yielded only a modest benefit at best; thus, nicotine alone may not be the driving factor.[35] Other proposed mechanisms have included modulating cellular immunity,[38] altering cytokine levels,[39] modifying colonic mucus production,[40] and predisposing patients to microvascular thrombi.[35,41] Although the relationship between smoking and IBD is well documented, a paradox exists in that the highest incidence of smoking is found in countries with the lowest incidence of IBD. For example, the incidence of IBD is higher in Canada[42] compared to South Korea[43]; however, the prevalence of smoking in Canada (22%)[44,45] is lower than in South Korea (65%).[45]

A similar relationship has also been proposed with passive smoke exposure.[32,34] However, a meta-analysis did not demonstrate an association for childhood passive smoke exposure or prenatal smoke exposure.[32] A possible explanation for these findings could be the presence of a dose-response relationship between smoking and IBD,[32] in which passive smoking may represent a lower level of exposure, leading to nonsignificant associations.[32]

Oral Contraceptive Pills

In 1995, a meta-analysis of 2 cohort studies and 7 case-control studies suggested that OCPs may marginally increase the risk of developing CD, but not UC.[46] This paper was limited by small sample sizes, and only 1 of the 9 included studies had a significant association. Additionally, the meta-analysis did not investigate a dose-response effect, as the majority of studies were performed before the introduction of lower OCP estrogen preparations.[47] A meta-analysis conducted in 2008 demonstrated a positive association for both UC and CD.[47] The risk of CD increased with prolonged exposure to OCPs, and its effect was reversed after the medication was discontinued.[47] Furthermore, upon investigating the potential for a dose-response effect, a reduction in the estrogen dose did not reduce the risk of CD; however, due to the limitations of the small sample size, similar investigations were not performed for UC.[47]

OCPs may increase the risk of developing IBD through the effects of estrogen. Estrogen acts as an

Table 1. Environmental Risk Factors for Inflammatory Bowel Disease

Risk factor	Ulcerative colitis	Crohn's disease
Smoking		
Current smoker	–	+
Ex-smoker	+	+
Never a smoker	+	–
Passive smoking: prenatal	Null?	Null?
Passive smoking: childhood	Null	Null
Appendectomy	–	?+
Oral contraceptive pills	+	+
Diet		
Sugars	?+	?+
Fats	?+	?+
Fruits and vegetables	?–	?–
Fiber	Null	?–
Breastfeeding	–	–
Infections/vaccinations		
Mycobacterium avium paratuberculosis	Null	?+
Measles infection	?Null	?Null
Measles vaccination	?Null	?Null
Adherent invasive *Escherichia coli*	?Null	?+
Psychrotrophic bacteria	?Null	?+
Perinatal infections	?+	?+
Antibiotics	?+	?+
Nonsteroidal anti-inflammatory drugs	?+	?+
Proxy measures of hygiene hypothesis		
Helicobacter pylori	?–	?–
Family size	?–	?–
Sibship	?–	?–
Birth order	?+	?+
Urban environment	?+	?+
Pets	?–	?–
Helminths	?–	?–
Prior gastroenteritis	?+	?+

immune enhancer, particularly in regard to humoral immunity and the proliferation of macrophages, whereas progesterone acts as an immune-suppressor.[48] 17-beta estradiol may effect tumor necrosis factor secretion and CD16 expression by macrophages.[48] Alternatively, estrogen may play a pathogenic role in IBD through a process of multifocal gastrointestinal infarction due to its thrombogenic potential.[47]

Appendectomy

Appendectomy is negatively associated with the development of UC, particularly among children experiencing appendicitis before 10 years of age. Meta-analyses investigating appendectomy and UC demonstrated a significant reduction in the risk of developing UC after an appendectomy.[49,50] In contrast, the relationship between appendectomy and CD is less clear.[51] Several studies have demonstrated that appendectomy is a risk factor for CD,[52-55] whereas other studies have shown an inverse association[56] or no association.[12,57-61] A meta-analysis demonstrated a significant risk of CD following an appendectomy.[51] However, a considerable proportion of the risk of developing CD was observed within the first year following an appendectomy, a time when incipient CD may lead to undue appendectomies.[51] After 5 years, the risk of CD was no longer significant, suggesting that a biological association between appendectomy and the development of CD is less likely.[51]

The mechanism by which appendicitis protects against the development of UC is not known; however, several hypotheses have been proposed.[50,62-64] The appendix may act as a reservoir of enteric bacteria and may be involved in antigen sampling that regulates the immunologic response to host microflora.[50,64] Furthermore, IBD is characterized by a shift in the balance toward a T-helper 1 cell-mediated inflammatory response in CD and a T-helper 2 response in UC.[65] A study by Andersson and colleagues suggests that appendicitis is mediated by T-helper 1 cells, which may explain the inverse associations between appendicitis and UC.[65]

Diet

Diet has been extensively studied in IBD[66]; however, these studies have yielded inconsistent findings.[67] A Japanese study found more than a 2-fold increased risk of CD following the consumption of sugars/sweeteners, sweets, fats and oils, and total fat.[68] Despite genetic susceptibility differences between Japan and the Western world (eg, low-prevalence *NOD2* variants in Japan), similar associations were found in North America.[69] In a Canadian study,[69] CD was associated with greater consumption of total fats, as well as monounsaturated and saturated fats; however, a negative relationship was found for carbohydrate consumption.[69] Similar associations have been found between UC and monounsaturated and polyunsaturated fat consumption.[70] Consumption of long-chain omega-3 fatty acids has been demonstrated to play a role in IBD[69]; however, findings remain inconsistent, with both protective[69] and risk[68,71] associations shown. Saturated and unsaturated fats may play a role in the inflammatory response through modulation of Toll-like receptors in macrophages.[72] High intake of dietary fiber, including fruit and vegetable consumption, has been shown to protect from IBD[3,68,69,73-75]; however, findings are inconclusive, as several studies have failed to find a relationship.[4,18,68] The mechanism by which fruits and vegetables confer protection may be related to their ability to modify enzymes involved in clearing reactive oxygen species.[69] However, CD patients with underlying strictures may avoid fiber to minimize symptoms, which would result in a biased association. Overall, most dietary studies have reported inconsistent findings, which highlights the challenges of studying diet (eg, recall bias) and the complex effects of diet on IBD development.

Breastfeeding

Breastfeeding, which protects infants against many other immune-mediated diseases,[76] may also reduce the risk of developing IBD. Although several studies support a protective role between breastfeeding and IBD,[76-79] other studies have failed to find an association,[22,74,80] while still others have shown a positive relationship.[18,81] A meta-analysis of 14 case-control studies found a protective role for breastfeeding in both CD and UC.[76] The mechanism by which breastfeeding may have a beneficial effect is likely multifactorial. Breastfeeding is important for acquiring oral tolerance to microflora and food antigens,[10,76] which may prevent IBD development.[76] Infant formula lacks lactoferrin, which is found in breastmilk[10] and may have antibacterial and antiviral effects,[82] as well as anti-inflammatory properties.[82] Although the meta-analysis found a negative association between breastfeeding and IBD, subsequent studies have produced conflicting evidence in which breastfeeding was found to be a significant risk factor for pediatric CD.[18]

Antibiotics

Exposure to antibiotics in childhood is hypothesized to interfere with the normal process of developing tolerance to enteric bacteria, which may lead to IBD.[83] Card and coworkers demonstrated a positive association between antibiotic use and the development of CD.[84] This finding is supported by evidence from other studies demonstrating a similar relationship.[74,83]

Nonsteroidal Anti-Inflammatory Drugs

A case-control study by Felder and colleagues investigating the effects of NSAIDs on IBD found a positive

association for both UC and CD.[85] NSAIDs can cause damage to the intestinal mucosa of the stomach, small bowel, and colon.[85,86] NSAIDs can also increase intestinal permeability by inhibiting cyclooxygenase, which reduces prostaglandin production.[85,86] Inhibition of prostaglandins has been implicated in IBD due to immunoregulatory effects,[87] particularly through the inhibition of tumor necrosis factor and the induction of anti-inflammatory cytokines such as interleukin (IL)-10.[87]

Microorganisms

Many microorganisms have been considered as possible causes of IBD. Several candidate organisms have been proposed in the pathogenesis of IBD, including *Mycobacterium avium* subspecies *paratuberculosis* (MAP), the measles virus, and adherent-invasive strains of *Escherichia coli* (AIEC).

Mycobacterium avium paratuberculosis

MAP has been postulated to cause CD due to the similarities between Johne's disease and CD.[3] Johne's disease manifests as a granulomatous ileitis in ruminants and is caused by MAP.[88] The transmission of MAP from infected animals to humans occurs through a variety of sources, including fecal shedding, contaminated raw meat, and consumption of raw milk.[5] Isolation of MAP in CD is inconsistent,[89-94] which is likely due to differences in methodologies. A study by Graham and colleagues[95] did not identify a relationship between the presence of *Mycobacterium* and CD; however, as MAP is slow-growing and does not thrive in standard culture conditions, use of this method may make detection of MAP difficult.[96] Using both polymerase chain reaction (PCR) and culture to test for MAP, Naser and associates[94] detected MAP in 46% of CD cases, 45% of UC cases, and 20% of controls. A study using a laser capture microscope to isolate subepithelial granulomas detected MAP DNA in 40% of examined CD cases and 0% of controls.[96] Another study using human intestinal mucosal biopsy specimens detected MAP in 92% of patients with CD and 26% of controls.[97] However, using PCR techniques, Bernstein and coworkers[92] did not identify a significant difference in MAP in biopsy samples between CD, UC, and healthy controls. Furthermore, a population-based matched case-control study demonstrated no differences in the rates of serology for MAP between CD, UC, and randomly sampled controls.[93] A meta-analysis of 28 case-control studies investigating the relationship between MAP and CD found a positive association when either enzyme-linked immunosorbent assay (ELISA) or PCR techniques were used to detect infection.[98] This meta-analysis included only ELISA and PCR studies, excluding studies conducted through culture methods. PCR studies primarily use the target sequence IS900 to detect MAP; however, this sequence can be found in other mycobacteria, resulting in reduced specificity of this technique.[99]

The presence of MAP in a subset of CD patients may indicate that MAP infection results in CD; alternatively, the development of CD may predispose benign colonization of MAP. Furthermore, CD occurs more commonly in urban centers. In contrast, rural regions and farmers have greater exposure to MAP, but increased rates of CD have not been observed in these populations. Treatment of MAP was not associated with long-term remission in a large double-blinded, placebo-controlled trial.[100] Furthermore, immune suppression has not been associated with widespread MAP infections, as observed with the use of biologics and tuberculosis. Consequently, the exact relationship between MAP and CD remains elusive.

Measles Virus

Paramyxoviral infection, particularly from the measles virus, has been explored in the pathogenesis of IBD.[10] The measles virus may persist in the mesenteric microvascular endothelium, leading to a chronic granulomatous vasculitis consistent with CD.[101,102] The measles virus has been identified in the endothelium, lymphocytes, and macrophages of inflammatory foci in CD patients that were not found in controls.[101,102] However, other studies have failed to confirm these findings using PCR techniques.[103-105] Epidemiologic studies have investigated this relationship, with similar inconsistencies. Ekbom and associates[106,107] reported an association between both prenatal and in-utero exposure to measles and CD later in life. However, subsequent studies have failed to find similar results.[108-111] The live attenuated measles vaccine has also been explored as a risk factor for IBD. Although one study demonstrated a relationship between the measles vaccine and IBD,[112] this association has not been reproduced in subsequent studies.[113-115] Consequently, the available evidence does not support an association between IBD and measles infection or measles-containing vaccination.

Helminths

IBD manifests in societies with reduced infestation of helminths. For the most part, the prevalence of IBD is inversely associated with the prevalence of helminth colonization. Helminths are thought to play an important immunoregulatory role with the intestinal flora.[10,116] Furthermore, open-label clinical trials of helminth treatment have shown a potential benefit for both UC and CD, which is likely secondary to the ability of the parasite to upregulate immunoregulatory Th2 cytokines (eg, IL-10, IL-4).[117-119]

Other Microorganisms

Several theories have proposed that IBD develops through dysbiosis between harmful and protective bacteria. Individuals diagnosed with an acute gastroenteritis have been shown to subsequently have an increased risk of developing IBD.[120] Pathogenic bacteria that cause gastroenteritis (such as *Salmonella* and *Campylobacter*) may play a role in the etiology of IBD.[121] AIEC has been shown to be specific for ileal CD and can invade intestinal epithelial cells and replicate within macrophages.[122] CD patients with *NOD2* variants, which predispose them to ileal CD, have been shown to have a reduced cytokine response to AIEC.[123] Defects in autophagy (eg, the *ATG16L* gene) may impair clearance of AIEC, leading to CD.[124] Alternatively, increased utilization of refrigeration (ie, cold chain hypothesis) has allowed psychotropic bacteria such as *Listeria monocytogenes* and *Yersinia enterocolitica* to thrive in modern societies. Exposure to these organisms has been theorized to increase the risk of developing IBD.[10,116] *Candida albicans*, a pro-inflammatory opportunistic pathogen, has been proposed to play a role in IBD; however, findings remain inconsistent.[125-127] This is likely due to the differing techniques used to detect the fungus (eg, ELISA, PCR) and the inherent challenges with these methods.

Stress

Psychological stress has been suggested to play a role in the etiology and pathogenesis of IBD due to the chronic, relapsing, and remitting nature of this disease.[128,129] Both chronic and acute stress can alter immune function.[128] Results from observational studies have been inconsistent, with findings supporting both positive and null associations.[128,129] However, due to the retrospective nature of these studies, recall bias may have influenced the results.[128] Evidence from animal models indicates that chronic psychological stress may exacerbate IBD by promoting damage to the intestinal mucosa, thereby impeding barrier function.[130,131]

Summary

Despite years of investigation, the environmental risk factors that have been identified have not explained the pathogenesis of IBD. Several environmental factors, such as smoking, appendicitis, OCPs, diet, breastfeeding, infections/vaccinations, antibiotics, helminths, and childhood hygiene, have been implicated in the increased worldwide incidence of IBD. However, even the most consistently demonstrated environmental risk factor, smoking, contributes only partially to disease pathogenesis (ie, most smokers do not have CD and most CD patients do not smoke). Thus, further studies are necessary to better understand the environmental determinants of IBD.

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