Enteroendocrine Cells: Sensing Gut Microbiota and Regulating Inflammatory Bowel Diseases

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Host sensing in the gut microbiota has been crucial in the regulation of intestinal homeostasis. Although inflammatory bowel diseases (IBDs), multifactorial chronic inflammatory conditions of the gastrointestinal tract, have been associated with intestinal dysbiosis, the detailed interactions between host and gut microbiota are still not completely understood. Enteroendocrine cells (EECs) represent 1% of the intestinal epithelium. Accumulating evidence indicates that EECs are key sensors of gut microbiota and/or microbial metabolites. They can secrete cytokines and peptide hormones in response to microbiota, either in traditional endocrine regulation or by paracrine impact on proximal tissues and/or cells or via afferent nerve fibers. Enteroendocrine cells also play crucial roles in mucosal immunity, gut barrier function, visceral hyperalgesia, and gastrointestinal (GI) motility, thereby regulating several GI diseases, including IBD. In this review, we will focus on EECs in sensing microbiota, correlating enteroendocrine perturbations with IBD, and the underlying mechanisms.

Key Words: enteroendocrine cells, gut microbiota, inflammatory bowel disease, immune system, gut dysfunction

INTRODUCTION

Inflammatory bowel diseases (IBDs), including ulcerative colitis (UC) and Crohn's disease (CD), is a group of chronic, recurrent, and relapsing diseases in the human gastrointestinal tract characterized by chronic intestinal inflammation and extra-intestinal manifestations.[1] The incidence of IBD has been increasing worldwide in recent years, consequently leading to an economic burden to both patients and society.[2, 3] Etiological studies of IBD have proposed several factors contributing to the occurrence of IBD, including the altered gut microbiota, abnormal host immune responses, genetic predispositions, and environmental factors.[4, 5] Enteroendocrine cells (EECs) represent around 1% of the total intestinal epithelium. Accumulating studies indicate that interactions between EECs and the gut microbiota may play an essential role in the pathogenesis of IBD. In this review, we will present an overview of the latest advances for roles of EECs in maintaining intestinal homeostasis. A comprehensive understanding of the functions of EECs would broaden our understanding of the mechanisms underlying the pathophysiology of IBD, and could thus identify potential therapeutic options for treatment of these diseases. For readers who are interested in the roles of enteroendocrine cells in inflammation beyond IBD, please read the elegant review article by Worthington et al.[6]

ENTEROENDOCRINE CELLS

Enteroendocrine cells are dispersed as single cells along the gastrointestinal mucosa in the crypts and villi and represent the largest endocrine system of the human body (Fig. 1).[7, 8] Traditionally, EECs can be divided into more than 10 different cell types based on their major secretory hormones (Table 1), including members of the chromogranin/secretogranin family, serotonin (5-HT), somatostatin, neuropeptide Y (NPY), vasoactive intestinal peptide (VIP), substance P (SP), cholecystokinin (CKK), glucagon-like peptide (GLP)-1/2, and Ghrelin.[9, 10] Of note, enterochromaffin cells, the largest population of EECs, can synthesize and secret 5-HT, accounting for >95% of the whole body's 5-HT.[11] Emerging evidence also demonstrates that several peptide hormones are likely to be co-expressed in the same EECs.[12, 13] For instance, enteroendocrine L-cells have been shown to express glucose-dependent insulinotropic peptide (GIP), cholecystokinin (CCK) neurotensin, and somatostatin in addition to GLP and peptide YY (PYY).[14, 15] The hormones produced by EECs may play essential roles in the regulation of nutrient absorption, intestinal immune response, epithelial barrier defense, visceral hyperalgesia, and colonic motility.[16] Chromogranin-A (CgA) is a heat-stable and soluble protein, which is stored and released from storage granules in the EECs.[17] Within the gut, several CgA-derived peptides (CgDPs) are generated by proteolytic cleavage of CgA (Table 2), such as vasostatin (VS), catestatin (CST), chromofungin (CHR), pancreastatin (PST), and serpinin.[18–20]

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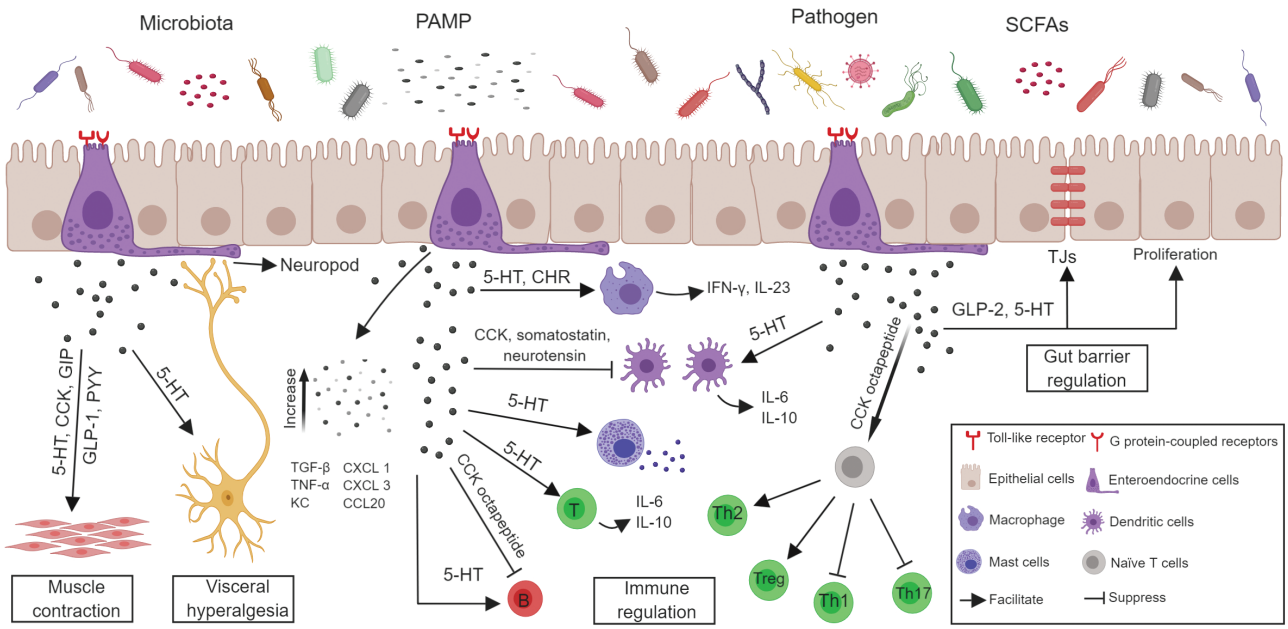

FIGURE 1. Enteroendocrine cell influence on gut function in inflammatory bowel disease. Enteroendocrine cells possess multiple chemosensory receptors that can detect intestinal microbiota and microbial metabolites. In response, EECs can secrete peptide hormones and classical cytokines to the surrounding immune cells and modulate both the innate and adaptive immune systems. Besides that, EECs possess cytoplasmic processes in close proximity to enteric nerve terminals. The released hormones may regulate the visceral hyperalgesia and intestinal motility in a paracrine fashion, along with synaptic transmission. Furthermore, enteroendocrine hormones can modulate the intestinal epithelial barrier function through both transcellular and paracellular pathways. All this evidence predicts the crucial role of EECs in the pathophysiology of IBD.

TABLE 1. EEC Cell Subtypes Based on Major Secretary Hormones

Cell Types	Major Secretory Hormones	Digestive Function
A (X-like) cell	Ghrelin, nesfatin-1	Appetite control, growth hormone release
G cell	Gastrin	Acidity, GI motility
D cell	Somatostatin	GI hormone release, GI motility, mucosal immunity
L cell	GLP-1, GLP-2, PYY	Appetite control, GI motility, energy homeostasis
K cell	GIP	Insulin secretion
I cell	CCK	Appetite control, GI motility, bile acid and digestive enzyme release, mucosal immunity
Enterochromaffin cell	5-HT	Appetite control, GI motor and secretory function, mucosal immunity
N cell	Neurotensin	GI motility, mucosal immunity
M cell	Motilin	GI motility
S cell	Secretin	Acidity, body fluid homeostasis
Enterochromaffin-like cell	Histamine	Acidity, mucosal immunity
P cell	Leptin	Appetite control, nutrients absorption, mucosal immunity

Enteroendocrine cells have specialized sensory microvilli reaching to the lumen and respond to intestinal luminal nutrients and/or microbiota by releasing hormones in a classical endocrine fashion. Further, morphological studies have revealed that several long pseudopod-like basal processes named "neuropods" extend from EECs, and the terminal end of these processes often resembles a synapse, which seems to interface with intestinal neurons or epithelial cells.[21, 22] These studies suggest that the EECs may affect neighboring cells or neurons by exocytosis of biological mediators through a paracrine effect, or by directly activating the afferent synaptic transmission.[23, 24]

GUT MICROBIOTA–ENTEROENDOCRINE CELL AXIS

The gastrointestinal microbiota consists of a group of microorganisms (bacteria, viruses, and some eukaryotes), with concentrations of up to 10^{11}–10^{12} cells/g luminal contents.[25]

TABLE 2. Physiological Functions of Chromogranin-A and its Derived Peptides in Intestinal Homeostasis

Molecule	Physiological Functions
CgA	Hormone and/or neuropeptide storage and release, gut microbiota composition, mucosal immunity, Ca^{2+} homeostasis
VS	Regulate intestinal permeability, repair intestinal mucosa, intestinal inflammation, GI motility
CST	Modulate leptin signaling, intestinal inflammation
CHR	Regulate GI motility, intestinal inflammation, epithelial tight junctions
PST	Regulate insulin secretion, gastric acid secretion, intestinal inflammation
Prochromacin	Antimicrobial activities
WE-14	Regulate intestinal inflammation
Parastatin	Regulate inflammation through vitamin D metabolic pathway
Serpinin	Regulate plasmin-induced inflammation and decrease cell death

Abbreviation: WE-14, a neuropeptide derived from the post-translational processing of CgA.

A healthy gut is colonized by >500 different microbial species.[26] The microbiota has established a harmonious ecosystem within the human organism, which contributes to the maintenance of normal immunological functions, facilitates nutrient digestion and absorption, prevents growth of pathogenic bacteria, and produces a variety of biologically important compounds, such as short-chain fatty acids (SCFAs), including principally acetate, propionate, and butyrate.[27, 28] Within the gut, acetate and propionate are mainly produced by bacteria of the *Bacteroidetes* phylum, whereas butyrate is generated by those of the *Firmicutes* phylum.[29] Notably, *Akkermansia muciniphila*, which is a mucin-degrading bacterium located in the colonic mucus layer, has been shown to produce SCFAs such as acetate and propionate.[30]

For the last 2 decades, IBD has been one of the most extensively investigated inflammatory disorders that are closely related with the altered gut microbiota. Increasing evidence has shown that reduced intestinal microbiota biodiversity and microbial dysbiosis appear to be important factors in the pathogenesis of IBD.[31–34] Large-scale gut microbiome sequencing associated with IBD reveals an increased prevalence of proteobacteria, including *Escherichia coli* and *Shigella,* and a significant decline of known beneficial bacteria, such as *Firmicutes, Enterobacteriaceae, Bacteroidetes, Roseburia intestinalis,* and the *Clostridium* XIVa and IV groups.[31–35] Further, an increased number of the genus *Fusobacterium* has been reported in the colonic mucosa of patients with UC.[36] Additionally, decreased butyrate-producing bacteria such as *Faecalibacterium prausnitzii* have been shown in IBD patients, which may account for the decreased amount of SCFAs in fecal samples from these patients.[37]

One potential mechanism underlying the interaction between the gut microbiota and IBD is microbial endocrinology, in which the bacteria regulate neuroendocrine hormone production in the host, which has been a focus of intense interest in recent years. Enteroendocrine cells express a vast array of receptors that play key roles in gut sensing (Fig. 2). Enteroendocrine cells express G protein-coupled receptors (GPCRs), including GPR40, GPR41, GPR43, GPR119, and GPR120, which have been identified as sensing receptors for gut microbiota-derived SCFAs,[38] or long-chain fatty acids (LCFAs) from triglyceride metabolites by pancreatic lipase digestion.[39] Both in vitro and in vivo studies demonstrate that EECs also express functional Toll-like receptors (TLRs; eg, TLR1, TLR2, TLR4, etc.) and respond to intestinal bacterial TLR ligands,[40] indicating that EECs might play an essential role in immune surveillance of luminal contents. Taste receptors (T_1Rs and T_2Rs, etc.) can respond to intestinal nutrients and beneficial compounds and play critical roles in nutrient assimilation and regulation of glucose homeostasis.[41, 42] EECs respond to bacterial quorum sensing molecules called acyl homoserine lactones from Gram-negative bacteria through activation of taste receptor type 2 member 38 (T_2R38).[43] *Clostridium sporogenes* expresses tryptophan decarboxylases to generate tryptamine, which is able to stimulate the production of 5-HT from enterochromaffin cells and modify the whole-body homeostasis.[44] In the gut mucosa of CD patients, enterochromaffin cells exhibit elevated transcripts for tryptophan hydroxylase 1 (TPH1) and TLR4, and *E. coli* lipopolysaccharides (LPS) stimulates more 5-HT release from the enterochromaffin cells of CD patients than those of healthy volunteers.[45] A concentration-dependent inhibitory effect of 5-HT on the *A. muciniphila* has been clarified using Tph1 gene knockout mice, indicating the role of 5-HT in regulating the gut microbiota and altering susceptibility to DSS-induced colitis.[46] CgA levels in EECs have been associated with gut microbial composition and diversity, in which the strongest association to CgA is observed from the *Archaea* species *Methanobrevibacter smithii*, whereas a negative association was reported from the phylum *Bacteroidetes.*[47] Furthermore, CgA levels in EECs have been considered a biomarker for colitis activity and response to therapy in patients with IBD.[48] Isovalerate, a microbial metabolite, can activate olfactory receptor 558 (Olfr558), which

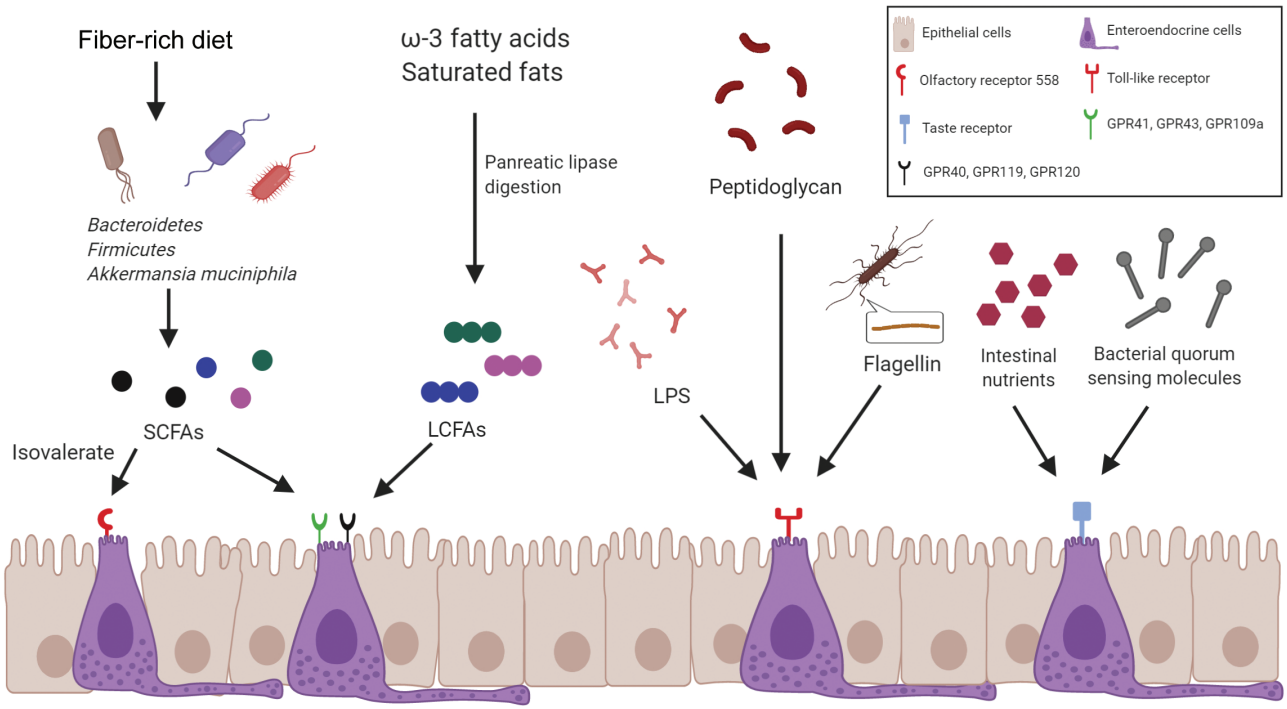

FIGURE 2. Interactions between the various receptors of enteroendocrine cells and the intestinal microbiota and/or microbial metabolites. G protein-coupled receptors, including GPR40, GPR41, GPR43, GPR119, and GPR120, have been recognized as sensing receptors for gut microbiota–derived SCFAs or LCFAs. Olfactory receptor 558, a microbial metabolite detector on EECs, can sense isovalerate, which is an SCFA. Toll-like receptors, such as TLR1, TLR2, and TLR4, can detect multiple intestinal bacterial TLR ligands (eg, bacterial LPS, flagellin, peptidoglycan). Taste receptors can respond to intestinal nutrients and bacterial quorum sensing molecules from Gram-negative bacteria.

is a microbial metabolite detector on enterochromaffin cells, and lead to voltage-gated Ca^{2+} channel-dependent 5-HT release from these cells.[49] Flagellin and bacterial LPS may act on the Toll-like receptor of EECs to enhance the inflammatory status associated with human IBD.[50]

ENTEROENDOCRINE CELLS IN IBD

Inflammatory bowel disease is characterized by a dysregulated immune response and inflammation-mediated mucosal damage, and its clinical manifestations include abnormal mucosal cytokine secretion, visceral hyperalgesia, and motility disorder. Paired-like homeobox 2b (*Phox2B*) and ubiquitination factor E4A (*UBE4A*),[51, 52] 2 enteroendocrine markers identified by genome-wide association studies, are increased in the terminal ileal tissue of CD patients, indicating a critical role for EECs in the pathogenesis of intestinal inflammatory disorders.[53] Alterations in enteroendocrine cell numbers and hormone secretion in the intestine have been demonstrated in both patients with IBD and animal models of colitis. By immunohistochemistry, the number of 5-HT-immunoreactive cells in the colon was significantly increased in both UC and CD patients.[54] Meanwhile, the densities of colonic PYY, pancreatic polypeptide (PP), and oxyntomodulin-producing endocrine cells were decreased in CD patients.[54] Consistent with IBD patients, the percentages of 5-HT-positive enterochromaffin cells were also increased in the inflamed ileum of guinea pigs suffering from trinitrobenzene sulfonic acid (TNBS)–induced colitis.[55] Additionally, the densities of 5-HT and oxyntomodulin-producing endocrine cells were increased, whereas PP production was decreased in the colon of rats after treatment with TNBS or dextran sulfate sodium (DSS).[55, 56] However, intestinal levels of CgA were increased in rats after administration with DSS but were decreased in the model of TNBS-induced colitis,[55, 56] indicating distinct EEC hormone responses to colitis under different triggers and pathogenic factors. Furthermore, altered circulating neuroendocrine synthesis and release have also been described in IBD. Patients with IBD exhibited elevated serum and plasma CgA,[57] whereas elevated fecal CgA levels were only found in patients with UC, but were not related to disease activity.[58] CgA and its derived peptides (eg, VS, CST, CHR, and chromacin) have been shown to participate in regulating antimicrobial activity, suggesting that altered CgA production in EECs may contribute to alterations in intestinal microbial composition, diversity, and functional richness.[47, 59] Significant alterations have also been demonstrated in other circulating EEC secretory products, such as PYY, CCK, GLP-1, 5-HT, somatostatin, gastrin, and motilin during the course of IBD.[60–66] Furthermore, neutrophil gelatinase-associated lipocalin (NGAL), a potential antimicrobial glycoprotein in

human neutrophils, has been found to be expressed in EECs in the healthy gut and in CD.[67]

MICROBIOTA–EECS AXIS IN THE REGULATION OF MUCOSAL IMMUNITY IN IBD

It has been well established that IBD is a result of an inappropriate mucosal immune response to gut microbiota, leading to chronic immune activation.[68] Among multiple levels of immune regulation that have been implicated in the pathogenesis of IBD, EECs, acting as the first line of pathogen detection, can secrete classical inflammatory cytokines and peptide hormones and have been widely studied in multiple inflammation- and infection-driven diseases of the gut.[6, 69, 70] In recent years, EECs have increasingly become of particular interest in understanding the pathogenesis of IBD.

Intestinal immune cells can express various receptors for secreted hormone peptides from EECs, including 5-HT, CCK, GLP-1, GLP-2, and neurotensin.[71] For instance, the expression of 7 isoforms of 5-HT receptor has been confirmed in mast cells, monocytes, dendritic cells (DCs), lymphocytes, and neutrophils.[72] The role of 5-HT as an immunomodulatory factor has been well established, including activating macrophages,[73] inducing proliferation of lymphocytes,[74] protecting natural killer (NK) cells from oxidative damage,[75] and promoting recruitment of T cells.[76] The amelioration of TNBS-induced colitis in monocyte chemoattractant protein-1-deficient (MCP-$1^{-/-}$) mice is associated with decreased infiltration of $CD3^+$ T cells and macrophages, along with decreased 5-HT-expressing enterochromaffin cells in the colonic mucosa.[77] $Itgb7^{tm1Cgn}$($\beta7^{-/-}$) mice, which lack natural gut intraepithelial T lymphocytes (natural IELs), were resistant to cardiovascular disease through GLP-1 production.[78] Several CgA-derived peptides have been described in the pathophysiology of intestinal inflammation of IBD. Chromofungin activates neutrophils and regulates the functions of macrophages in the colon through an NF-κB dependent pathway.[79, 80] Pancreastatin can upregulate the expressions of pro-inflammatory mediators, including interleukin (IL)-1β, IL-6, tumor necrosis factor (TNF)–α, IL-12p70, and interferon (IFN)-γ,[81] and downregulate anti-inflammatory cytokines such as IL-10.[82] Catestatin has been demonstrated to have a beneficial effect against intestinal inflammation via modulation of the innate immune cells and gut microbial composition.[83] Accumulating evidence indicates a significant correlation between the abnormalities of intestinal endocrine cells and the mucosal recruitment of activated cells from both the innate and adaptive immune systems in IBD.[55, 84–88]

Innate immune cells, such as macrophages and DCs, can sense the intestinal microbiota and initiate the innate immune response, which represents the first line of defense against the microorganisms of the intestinal flora. Higher frequencies of macrophages and DCs are present in the intestinal inflammatory lesions of human IBD.[89–93] In the inflamed mucosa of CD patients, macrophages are increased and produce large amounts of pro-inflammatory cytokines (IFN-γ, IL-23) in response to commensal bacteria, such as *Escherichia faecalis*.[90] Carboxypeptidase E (CPE), an enzyme critical for the synthesis of most hormones, is specifically expressed in EECs. Deficiency of CPE in mice leads to increased migration of myeloid macrophages into the mucosa and subsequently an aggravated severity of DSS-induced colitis.[94] Higher levels of activated DCs have been found during a flare-up in IBD patients.[95, 96] Several EEC peptide hormones, including CCK, somatostatin, and neurotensin, are reported to be inhibitory to DC activation,[97–99] which might be beneficial for managing these patients. By binding to 5-HTR_3, 5-HTR_4, and 5-HTR_7 receptors, 5-HT regulates DCs to release pro-inflammatory cytokines, such as IL-6, and promote CCL22/MDC chemokine production.[100] In tryptophan hydroxylase-1-deficient ($TPH1^{-/-}$) mice, the decreased levels of 5-HT are accompanied by the reduced production of IL-12p40 by DCs, which accounts for the attenuation of colitis after exposure to DSS.[101] A positive correlation between mast cell number and the density of enterochromaffin cells has been found in the ileum of IBD patients,[102] and application of 5-HT receptor antagonists (methysergide or ketanserin) could reduce endotoxin-induced mesenteric mast cell activation in vivo, probably via the 5-HT(2A) receptor subtype.[103]

Enteroendocrine cells possess functional TLRs, which are recognized as crucial components of the innate immune response. In response to LPS stimulation, STC-1 cells, a murine EEC line, neutralize intestinal bacteria by releasing keratinocyte-derived chemokine and β-defensin 2[104] and promoting the expression of the proinflammatory cytokine TNF-α and the anti-inflammatory cytokine TGF-β through the activation of intracellular NF-κB and MAPK pathways.[105] Consistently, LCC-18 cells, a human enteroendocrine cell line, produce chemoattractant molecules (eg, CXCL 1 and 3 and CCL20) to recruit innate immune cells from the colonic lamina propria in response to stimulation of LPS and flagellin.[50]

The potential interactions between EECs and the surrounding adaptive immune cells in the pathogenesis of IBD remain largely unexplored. In the colonic mucosa of rhesus monkeys, 5-HT-positive EECs are in contact with, or in close proximity to, both $CD3^+$ T cells and $CD20^+$ B cells, which suggests a possible role of 5-HT in the regulation of intestinal adaptive immune responses.[106] Further investigations have demonstrated that 5-HT could directly act on T and B cells to regulate their activation and proliferation through its receptors expressed on lymphocytes.[107, 108] Cholecystokinin octapeptide can directly affect T cells and B cells. It has been shown that CCK octapeptide suppresses Th1 and Th17 differentiation but promotes Th2 and Treg development.[109, 110] Cholecystokinin octapeptide also inhibits the production of co-stimulatory molecules (CD80, CD86) in LPS-activated B cells.[109, 110] The presence of IL-13 receptor α1 on both 5-HT and CgA-expressing enteroendocrine cells further verifies the importance of the

immunoendocrine axis in the gut.[111] A better understanding of the interaction of EECs and immune cells in the regulation of the pathogenesis of IBD is required in future investigations.

MICROBIOTA–EECS INTERACTION IN THE REGULATION OF GUT BARRIER FUNCTION IN IBD

The intestinal epithelium, composed of intestinal epithelial cells (IECs), goblet cells, Paneth cells, tuft cells, and EECs, serves as the interface for digestion and nutrient absorption and is a critical defensive barrier against toxins and microorganisms. The intestinal barrier is regulated by a number of intercellular junctional complexes, including tight junctions (TJs). Accumulating evidence shows that epithelial morphological changes, including increased TJ breaks in IBD, contribute to an increase in intestinal epithelium permeability, which promotes abnormal translocation of the gut microbiota or gut microbiota components from the gut lumen to the host blood and tissues.[112, 113]

GLP-2, secreted from EECs in the intestine after food intake, is upregulated in patients with IBD.[114] It can act directly on human IECs to promote their proliferation[115] and wound healing through induction of epithelial cell migration mediated by TGF-β.[116] GLP-2 has also been demonstrated to decrease colonic crypt cell apoptosis, increase crypt depth and colon length, and protect colonic mucosal architecture in DSS-induced colitis.[117–119] Subcutaneous administration of GLP-2 in CD-1 mice decreases the intestinal conductance and unidirectional fluxes, indicating that GLP-2 treatment might improve intestinal barrier function by affecting both paracellular and transcellular pathways.[120] Furthermore, specific tight junction protein expression, such as ZO-1 and occludin, is upregulated in GLP-2-treated Caco-2 cells via a TNFα-mediated process.[121] Additionally, *Akkermansia muciniphila*, which resides in the mucus layer and plays a role in gut barrier function, has been shown to be decreased in patients with IBD.[122] Its effects on barrier function could be mediated by 2-oleoylglycerol (2-OG), which stimulates the secretion of GLP through enteroendocrine L cells.[123, 124]

Piezo2, a mechanosensitive ion channel, has been demonstrated to be expressed in a subset of human and mouse EECs.[125] Mechanical stimulation of these EECs leads to an intracellular Ca^{2+} increase, 5-HT release, and pressure-induced epithelial fluid secretion.[126] Furthermore, human enteric adenovirus 41 could infect enterochromaffin cells and induce the release of 5-HT,[127] which might activate enteric glia.[128] Furthermore, EECs in the *Drosophila* midgut could respond to the pathogenic bacterium *Pseudomonas entomophila* by expressing the prosecretory transcription factor *dimm*, which is necessary for the induction of antimicrobial peptides at the barrier epithelium.[129]

MICROBIOTA–EECS INTERACTION IN THE REGULATION OF VISCERAL HYPERALGESIA IN IBD

Visceral hyperalgesia or an increased sensation of physical stimuli in the gut is relatively common in patients with IBD. A correlation between visceral pain disorders and alteration of the intestinal microbiota or microbial products has been demonstrated in these patients. Inflammatory bowel disease patients show decreased numbers of butyrate-producing bacteria (eg, *Roseburia inulinivorans, Ruminococcus torques, C. lavalense, B. uniformis,* and *F. prausnitzii.*) and a subsequent reduction of butyrate levels in the gut, resulting in changes in visceral hyperalgesia.[130, 131] In germ-free mice, inflammatory hypernociception, which is induced by various stimuli, including LPS and IL-1β, is reduced.[132] Accumulating evidence indicates that EECs regulate visceral hyperalgesia in IBD.

Enteroendocrine cells communicate with the enteric nervous system (ENS) through hormone secretion.[133] Serotonin is a key neurotransmitter in control of nociceptive responses, with receptors located in the peripheral and central nervous systems. Increased secretion of 5-HT has been shown in the enterochromaffin cells of patients with IBD and experimental colitis models,[54–56, 134–136] which could stimulate the 5-HT_3 and 5-HT_4 receptors expressed on the primary afferent neurons of both the splanchnic and vagal fibers and correlate with the severity of abdominal pain.[137] Specific receptors on enterochromaffin cells, including transient receptor potential ankyrin 1 channel (TRPA1) sensing irritation, Olfr558 sensing microbial metabolites, and TRPC4, an α2A adrenoreceptor sensing stress-related catecholamines, are proposed to be involved in sensory transduction pathways by controlling 5-HT release.[49]

Using 3D reconstruction of confocal microscopic images, recent evidence demonstrates that EECs possess cytoplasmic processes termed neuropods, which are surrounded by glia and in close contact with enteric nerve terminals, including sensory nerve endings.[21, 22] These findings reveal the possibility of a novel neurotransmission mediating the responsiveness of EECs to the bacterial by-products in the lumen of the intestine. Isovalerate, a microbial metabolite, could bind to Olfr558 on enterochromaffin cells and consequently modulate $5HT_3R$-expressing primary afferent nerve fibers via synaptic connections.[49] Furthermore, using serial block face scanning electron microscopy, the neuropods of enteroendocrine cells are escorted by enteric glial cells, which have a similar morphology and function as astrocytes in the brain and have been shown to be involved in the inducing process of visceral hyperalgesia.[138] A recent study demonstrated that neuropod cells transduced glucose stimuli to vagal neurons by releasing the neurotransmitter glutamate, helping the brain make sense of the gut luminal signals rapidly.[139]

MICROBIOTA–EECS INTERACTION IN THE REGULATION OF GUT MOTILITY IN IBD

A change in gut motility has been found in symptomatic IBD, such as decreased segmenting colonic contractions and increased colonic propagating contractions, which may induce efficient anterograde movement of luminal contents,[140–142] resulting in symptoms of diarrhea, at least in UC patients.[141] Several enteroendocrine hormones, such as CCK, GIP, GLP-1, and PYY, are essential mediators of postprandial gastrointestinal motility.[143] SCFAs, acting through GRP41 and GPR43 on EECs,[144] could stimulate the release of 5-HT and subsequently provoke the secretion of acetylcholine from the colonic myenteric plexus, resulting in muscle contractions.[145] Indigenous spore-forming bacteria from the gut microbiota might regulate 5-HT biosynthesis in EECs through promoting TPH1 expression.[146] Non-neural pathways, such as the cyclo-oxygenase products prostaglandins, have been reported to be involved in the propionate-induced colonic tonic contraction through their direct actions on circular muscle.[147] TRPA1, activated by thermal nociception, natural plant-derived products, and inflammatory hyperalgesia, along with olfactory receptor,[148–150] has been shown to be expressed in enterochromaffin cells and contributes to the production of 5-HT.[151] The release of 5-HT could evoke intestinal contractions of isolated guinea pig ileum, which is significantly inhibited via the 5-HT_3 receptor antagonist.[152] *Drosophila* provides the ideal intestinal model system to investigate the functional implication of EECs in the context of the host–microbiota interaction. Peptide hormone Diuretic Hormone 31, expressed by a group of EECs, is responsible for peristalsis in the junction region of the *Drosophila* larval midgut.[153] The hypochlorous acid–sensitive receptor expressed in a subset of midgut EECs might facilitate enteric expulsion of the opportunistic pathogen *Erwinia carotovora* through activation of the Duox pathway, thus maintaining the microbiota's homeostasis.[154] These results indicate that the gut microbiota and enteroendocrine hormones might play pivotal roles in GI motility; however, the role of EECs in microbiota-induced gut motility disorders remains to be further clarified in the pathophysiology of IBD.

CONCLUSIONS

Inflammatory bowel disease is a chronic intestinal inflammatory disease affecting patients' quality of life. Accumulating evidence has demonstrated that the interactions of enteroendocrine cells and the gut microbiota contribute to the maintenance of intestinal homeostasis. Enteroendocrine cells, as firstline sensors for the microbiota, can secrete classical cytokines and hormonal peptides to directly and indirectly regulate several functions in different cell types in the GI tract. Their involvement in microbial sensing and their roles in the regulation of innate and adaptive immune responses, the intestinal epithelial barrier, visceral hyperalgesia, and gut motility emphasize the importance of EECs in the regulation of the pathogenesis of IBD. Investigation of the gut microbiota–EEC interaction in IBD will provide great insights into the pathogenesis of IBD and the development of tools for the prediction, diagnosis, and treatment of IBD.

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