Gut Microbiome and Its Role in the Pathophysiology of Irritable Bowel Syndrome

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Summary

Irritable bowel syndrome is the most common functional gastrointestinal disorder, affecting up to 9% individuals globally. Although the etiology of this syndrome is likely heterogenous, it presents with its hallmark symptoms of abdominal pain and altered intestinal motility. Moreover, it is considered to be a disorder of the gut-brain interaction, and the microbiome has often been implicated as a central player in its pathophysiology. Patients with irritable bowel syndrome display altered composition and function of the gut microbiota compared to healthy controls. Microbiome directed therapies, such as probiotics, antibiotics and fecal microbiome transplantation, appear to be beneficial for both gut symptoms and psychiatric comorbidities. This review aims to recapitulate the available literature on the microbiome contribution to the pathophysiology and symptoms presentation of irritable bowel syndrome, as well as the current literature on microbiome-targeted treatments for this disease.

Keywords. *IBS, diet, microbiome, FMT.*

El papel de la microbiota intestinal en la fisiopatología del síndrome de intestino irritable

Resumen

El síndrome de intestino irritable es el trastorno digestivo funcional más diagnosticado, el cual afecta hasta el 9% de la población mundial. Aunque la etiología y las manifestaciones clínicas de esta enfermedad son muy variables, se caracteriza por la presencia de dolor abdominal y alteraciones en la motilidad intestinal. Se considera un desorden del eje intestino-cerebro y se ha planteado que el microbioma intestinal juega un papel central en su fisiopatología. De hecho, los pacientes diagnosticados con síndrome de intestino irritable presentan alteraciones en la composición y función de la microbiota intestinal en comparación con controles sanos. En línea con esta hipótesis, varios estudios confirman que pacientes con este trastorno pueden beneficiarse, tanto a nivel gastrointestinal como psicológico, de intervenciones dietéticas y del uso de terapias dirigidas al microbioma como son el uso de probióticos, antibióticos y, más recientemente, del trasplante fecal. El objetivo de este artículo es llevar a cabo una revisión bibliográfica de la evidencia científica que apoya el papel de la microbiota en la fisiopatología y sintomatología de síndrome del intestino irritable, así como el uso de enfoques terapéuticos dietéticos o microbianos para el tratamiento de pacientes con esta enfermedad.

Palabras claves. *SII, dieta, microbioma, transplante de materia fecal.*

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Abbreviations

IBS: Irritable bowel syndrome.

GI: Gastrointestinal.
IBS-C: Irritable bowel syndrome with constipation.
IBS-D: Irritable bowel syndrome with diarrhea.
ENS: Enteric nervous system.
5-HT: 5-hydroxytryptamine.
AHR: Aryl-hydrocarbon.
PI-IBS: Post-infectious IBS.
SIBO: Small intestinal bacterial overgrowth.
FMT: Fecal microbiome transplant.
FODMAPs: Fermentable oligo-, di- and monosaccharides and polyols.
SCFAs: Short chain fatty acids.
RCT: Randomized controlled trial.

Introduction

Irritable bowel syndrome (IBS) is the most common functional bowel disorder worldwide affecting between 5% and 10% individuals globally.[1, 2] Its prevalence varies across the world according to the diagnostic definition used (Rome III vs. Rome IV), the population selected, and local factors.[2] As there is no diagnostic biomarker for IBS, its diagnosis is based on symptom reporting. Rome IV is the latest iteration of the Rome Diagnostic criteria and with its rather strict approach, the global prevalence of IBS was found to be lower (4.1% pooled prevalence) than the one previously reported using the Rome III criteria (10.1% pooled prevalence).[3] IBS is one of the most common reasons of healthcare seeking with significant socioeconomic impact.[1]

Despite being the most studied functional gastrointestinal (GI) disorder, its pathophysiology is incompletely understood, in part due to many factors involved in its genesis. It is now well accepted that IBS is a disorder of the gut-brain communication, presenting with visceral hypersensitivity, intestinal dysmotility, impaired central processing of stimuli arising from the GI tract, altered gut microbiota, as well as frequent psychiatric comorbidities, such as anxiety and depression. In addition, multiple dietary triggers are commonly reported by IBS patients.

IBS can be classified into 4 different categories according to bowel habits and stool form: IBS with constipation (IBS-C), IBS with diarrhea (IBS-D), Mixed IBS, and Unsubtyped IBS.[4] IBS affects patients across the lifespan, but there is an overall strong female predominance, modulated by age and hormonal status.[5, 6] Sex hormones also influence IBS symptom severity and subtype, with constipation being predominant in females, and diarrhea in males.[6, 7] While all these factors play a role in IBS pathophysiology, this review will focus on the role of gut microbiota and the possible use of microbiota-directed therapies for symptoms' mitigation.

Gut Microbiota and Healthy Gut

Mammals shelter in their body an incredibly complex and diverse community of microorganisms, collectively called microbiota (or microbiome if we refer to all the microorganisms and their genetic content), which comprises archaea, bacteria, viruses, fungi, and eukaryotes.[8, 9] The gut microbiota evolves during early life until a unique, subject-specific (fingerprint) adult-like community arises, which is highly resilient and relatively stable throughout life,[10-12] being dominated by few phyla only, mainly Firmicutes and Bacteroidetes, together with members of Actinobacteria, Verrucomicrobia, Proteobacteria, Fusobacteria, and Cyanobacteria.[13] Diet, regional variability and ethnicity greatly impact gut microbiota composition.[14, 15]

Gut microbiota is thought to be functionally redundant, meaning that different bacterial consortia perform similar functions in different individuals.[16] Indeed, gut microbiota carries out essential functions that the human body is unable to perform,[17, 18] while occupying a unique, nutrient rich niche. The central role of the microbiome is highlighted by studies in germ-free or microbiome depleted animals, which demonstrated that gut microbiota is required for normal gut physiology, metabolism, a balanced immune system,[19-25] regular development of the enteric nervous system (ENS),[26, 27] and a normal perception of inflammatory, mechanical and visceral pain.[28-30] In addition, gut microbiota affects GI motility[31-34] due to its effects on the ENS, by modulating the expression of toll like receptors, serotonin (5-HT) release and activation of the aryl-hydrocarbon (AHR) and the 5-HT_4 receptors.[26, 35-37]

The Role of Gut Microbiota in IBS

The implication of the gut microbiome in IBS is not a novel concept, as many studies have shown that infectious gastroenteritis is the most common trigger of IBS in previously healthy individuals.[38-41] Post-infectious IBS (PI-IBS) can develop immediately after a bacterial, viral or protozoal infection, or chronic GI symptoms can worsen after an infectious gastroenteritis (for a detailed review see Berumen *et al.*, 2021).[42] The underlying mechanisms are still to be fully elucidated, although several studies have shown evidences of low-grade inflammation or immune activation in IBS patients.[43] It has been proposed that transient inflammation leads to subtle but permanent changes in the structure and function of the gut, in-

cluding increased infiltration with lymphocytes and mast cells, altered enteric nerves and enterochromaffin cells, that, in turn, induce GI symptoms.[44]

The structure and function of the gut microbiota is deeply perturbed at the site of the infection[45] and it might act synergistically with ongoing inflammation and impaired epithelial permeability, increasing the risk of IBS development in susceptible individuals.[40, 42] Another condition that has been often associated with IBS is the small intestinal bacterial overgrowth (SIBO), which may be responsible for symptom generation in some patients with IBS. SIBO is defined as a quantitative alteration of the small intestinal microbiota (reviewed in Bushyhead & Quigley, 2021).[46] When the mechanisms in place to control and limit bacterial overgrowth in the small intestine (IgA secretion, gastric acid, bile acid and pancreatic secretions, as well as motor patterns) fail, pathological colonization occurs.[46] Bacterial overgrowth results in unusual fermentation with increases in gas production, abdominal bloating, malabsorption, abdominal pain, diarrhea, and abnormal GI motility.[46-49] There appears to be a link between SIBO and IBS;[50] however, its role in IBS is controversial, in part due to the scientific community not reaching a consensus on the detection method to use. While the breath tests are not well validated for SIBO, the jejunal aspirates are not always accurate.[50-54] Thus, it remains unclear whether SIBO is actually fundamental to the pathophysiology of IBS or is just a complicating phenomenon. However, several studies suggested that treatment of SIBO with non-absorbable antibiotics, rifaximin being the most commonly used, improves gut symptoms in a proportion of patients with IBS.[46, 55-58]

Alterations in the gut microbiota composition of IBS patients have been increasingly reported during the last decade, and while multiple studies have shown differences in the microbiota composition between IBS patients and healthy controls (recently reviewed by Pittayanon *et al.*, 2019, and Duan *et al.*, 2019),[59,60] the results of these studies have been inconsistent and no unique IBS bacterial signature or profile has been identified. This could be partially due to the use of different detection methods, as well as to different patient populations. In general, there appear to be an increase in potentially pathogenic bacteria, often facultative anaerobes or aerobes, with a decrease in strict anaerobic bacteria, and a decrease in bacterial diversity in IBS patients compared to healthy controls.[59, 61] It should be noted, however, that there is not a clear consensus on what constitutes a healthy microbiota.[62, 63]

A recent study reported that a great proportion of IBS patients (57% in this study) present with visible colonic biofilms, harbouring a less diverse microbiome with an overgrowth of *Escherichia coli* and *Ruminococcus gnavus* spp.[64] These biofilms correlated with an altered gut microbiome composition and with bile acids malabsorption.[64] While most IBS studies focused on the bacterial compartment of the gut microbiota, few studies that have researched its other components found differences in the mycobiome[65, 66] and the virome,[67] that were associated to those of the bacteriome.[66, 67]

However, it is now accepted that changes in microbial metabolic activity may have more impact on the host than the changes in microbial profiles. A recent study highlighted not only the importance of longitudinal sampling for IBS, given the fluctuating nature of IBS symptoms, but also the value in integrating different type of data, such as multiple-omics (metagenomics and metabolomics from host and microbes), as well as metadata on symptoms and gut physiology.[68, 69] Indeed, these studies found that IBS symptom severity fluctuates in parallel with functional variations in the gut microbiota, as well as with altered bile acid and purine metabolism.[69] Nevertheless, a unique and shared metabolomic dysfunction has yet to be discovered for IBS patients. Furthermore, it remains unclear whether the altered microbiome observed in clinical studies is a cause, or a consequence, of the gut dysfunction. Animal studies, employing gnotobiotic models in which germ-free mice are colonized with microbiota from patients with IBS or healthy controls, have proven the causal role of gut microbes in IBS pathophysiology, including altered motility, permeability, visceral hypersensitivity, immune activation, and psychiatric comorbidities.[70-73] Nonetheless, further mechanistic studies are needed to advance the field into personalized medicine and microbiota-targeted therapies.

Microbiota-Directed Therapies of IBS

The growing body of evidence suggesting the key role of bacteria in IBS has led to the design of many interventional studies targeting the gut microbiota of IBS patients. Unfortunately, results of these studies have been rather inconsistent. Microbiome targeted approaches include the use of dietary interventions, probiotics and prebiotics (already discussed in depth by Valdovinos-Díaz M.A. in the previous issue of this journal),[74] antibiotics, and, more recently, fecal microbiome transplantation (FMT). Dietary interventions are often a preferred, as non invasive, first line of treatement for IBS symptoms. Chronic diet is a major microbiome modulator, as our microbes eat what we eat. Dietary triggers, such as gluten or highly

fermentable oligo-, di- and monosaccharides and polyols (FODMAPs), have been frequently reported to worsen symptoms in IBS patients (60%).[75,76] Thus, dietary interventions are often proposed by clinicians or self-administered by patients as initial therapuetic approaches to curb IBS symptoms.

A systematic review for the American College of Gastroenterology found insufficient evidence to recommend excluding gluten to reduce IBS symptoms, due to paucity of randomized, placebo controlled trials, and a low quality evidence suggesting that reduction in FODMAPs intake reduces IBS symptoms.[76] However, a more recent meta-analysis found that low FODMAP diet is indeed more effective at reducing GI symptoms, such as abdominal pain and abdominal bloating, as well as improving quality of life, than traditional dietary advice or control diets.[77] Long-term effects of a low FODMAP diet, however, have been questioned, given that patients could develop nutritional deficits or detrimental loss of beneficial bacteria, such as Bifidobacteria.[78] Staudacher and colleagues have recently reported the results of the first long-term personalized low FODMAP study, in which Bifidobacteria levels were unaffected, but a significant decrease in short chain fatty acids (SCFAs) was observed.[79] The long-term consequences of this SCFAs impairment are unknown, and, as SCFAs have been implicated in regulation of GI motility and gut epithelial function,[80, 81] this observation requires further studies. This study exemplifies very well the great conundrum behind diet-microbiome-directed therapies, as modifying one dietary component may have a temporal beneficial effect while possibly triggering a long-term ripple effect due to microbiome restructuring.

Another microbiome-targeted approach that has been employed for the treatement of IBS symptoms is rifaximin: a minimally absorbed antibiotic normally used for SIBO and as second-line treatement for IBS-D.[58, 82] The evidence for its efficacy is, however, only modest.[82, 83] Besides rifaximin, two other antibiotics have been tested in IBS patients, neomycin and norfloxacin[83-85], with both medications being more effective than placebo at improving IBS symptoms[83-85]. However, repeated use of antibiotics in IBS is discouraged, and should not to be confused with its use for SIBO, as it could lead to increased microbial antibiotic resistance gene pool in IBS patients.

The last microbiome-targeted approach that has increasingly gained attention is the FMT. Seven randomized controlled trials (RCTs) have been performed up until now, with three of them reporting clear beneficial effects including reduction of IBS symptoms[86-88] and improving quality of life.[87, 88] Three RCTs found no clear efficacy of FMT in IBS[89, 90, 91] and one found only a transient relief of symptoms.[92] Two additional studies have investigated the long-term efficacy of FMT observing sustained efficacy[93, 94] and safety,[94] and changes to the gut microbiome that were more comprehensive that those observed at the end of the original RCT.[93] All these studies, however, did not use a standardized method of delivery, with some administering the FMT into the cecum or the distal colon,[86, 91, 92] some into the small bowel,[87, 88] and others using oral capsules.[89, 90] Similarly, these studies differed with the respect to the donors, with some using only one donor,[87, 92] some multiple donors[86, 88-91] and others pooling all donors together.[86, 89] Furthermore, patient preparation was not identical, with some studies using bowel preparation or prescribing loperamide as pre-treatment.[86, 88]

Despite these differences in the study design and outcomes, some preliminary conclusions may be reached: donor selection appears to be crucial, and pooled microbiota from several donors may have worse outcomes than that of single donor. The amount of material (> 30g) and frequency of administration seems to play a major role, with repeated FMT having better efficacy.[94, 95] Finally, donor microbiota engraftment does not appear to be necessary for the successful outcome. Based on gnotobiotic mouse models, the donor selection may be the key factor, as behavioral and physiological abnormalites seen in patients can be transferred into germ-free mice through microbiota transplantation,[73] thus highlighting not only the necessity to screen in depth potential donors for physical and mental health, but also the potential for treating psychiatric comorbidities of IBS.[96]

In conclusion, accumulating data suggest that, in a significant proportion of patients, the microbiota plays an important role in the genesis and maintenance of IBS. The use of personalized dietary approaches, probiotics and other microbiota directed therapies, including FMT, appears to be of therapeutic value, although more clinical data are needed. We should strive to bridge the gap currently existing between preclinical and clinical research[69] with further mechanistic translational and reverse translational studies to elucidate the complex interactions behind success and failure of these microbiome-directed therapies.

Intellectual Property. *The authors declare that the data that appear in this article are original and were made in their belonging institutions.*

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