Advances in the understanding of the intestinal micro-environment and inflammatory bowel disease

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Abstract
The human gastrointestinal tract accommodates an entire micro-environment for divergent physiologic processes, the dysbiosis of this micro-ecology has a strong inter-action with the pathogenesis of inflammatory bowel disease (IBD). In the past few years, with the advances in the understanding of microbiome, its metabolites and further application of next generation sequencing, analysis of dynamic alteration of gut micro-environment was realized, which provides numerous information beyond simple microbiota structure or metabolites differences under chronic colitis status. The subsequent intervention strategies targeting the modulation of intestinal micro-environment have been explored as a potential therapy. In this review, we will summarize the recent knowledge about multi-dimensional dysbiosis, the inter-action between fungus and bacteria under inflamed mucosa, and the clinical application of probiotics and fecal microbiota transplantation as a promising therapeutic approach in IBD.
Keywords: Intestinal micro-environment dysbiosis; Inflammatory bowel disease; Multi-omics; Probiotics; Fecal microbiota transplantation

Introduction

Inflammatory bowel disease (IBD) is a type of relapsing inflammatory disorder of the gastrointestinal tract, which involves ulcerative colitis (UC) and Crohn's disease (CD) that are closely related to genetic susceptibility, dietary influences, and the shift of host-microbiota inter-actions.[1] Although the main mechanism of pathogenesis remains unknown, the subtle alteration of microbiota configuration and related functional changes are supposed to participate in the pathogenesis, recurrence and drug resistance in this refractory disease. Therefore, it has become the emerging intervention targets for the therapeutic of IBD.[2] There is considerable interest in the intestinal micro-environment dynamic alteration in the relapsing colitis status and the potential mechanisms that may involve in the pathogenesis and refractoriness of IBD. This review highlights the inter-play of micro-organisms, multi-omics alteration, and the application of microbiota manipulation in this chronic immune-mediated, intestinal micro-flora dysbiotic inflammatory condition.

Over the past decade, our knowledge on the micro-environment has boomed, with the development of next-generation sequencing, especially the widely application of meta-genomics, 18S ribosomal RNA (rRNA) sequencing, viromics, and meta-bolomics, we could decipher more obscure and subtle microbiota configurative and functional alteration with higher efficiency and deeper magnitude that could not be obtained by traditional culture-dependent methods.[3]

Dysbiosis of the Micro-Environment in IBD

It is well accepted that the microbiota signatures of patients with IBD are distinctive to healthy individuals, which is known as dysbiosis [Figure 1]. However, there is not an accurate parameter that reflects the degree of dysbiosis, and the most widely used indirect index is microbial diversity. Several factors can destroy the relatively balanced intestinal micro-environment such as daily diet, antibiotics intervention, other oral medications, and even bowel preparation before colonoscopy.[4,5] Overall, host genetics, mucosal transcription level, and metabolic products contribute to the main part of species-based microbiota taxonomy, while disease status and diet only take up a small part of microbiota structure variation. Principal co-ordinate analysis based on Bray-Curtis dissimilarities of microbiota species was implemented to

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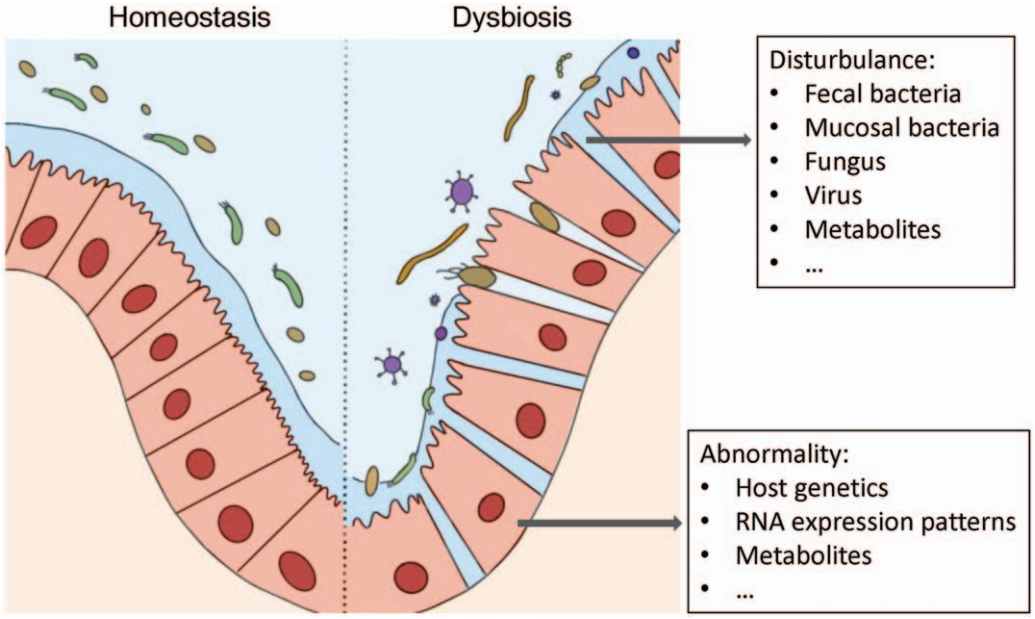

Figure 1: The intestinal micro-environment dysbiosis in multi-dimension.

quantize dysbiosis score, even non-IBD individuals can have a dysbiotic micro-environment and experienced pronounced repeated shift in microbiota structure in a period of time.[6]

Microbiota dysbiosis in patients with IBD, based on microbial composition alterations, has been confirmed in previous works,[7] including the decline of bacterial diversity and bacterial load. However, data focusing on functional changes of the intestinal community are still lacking. Recent findings have demonstrated that the disturbance of metabolites is associated with the pathogenesis of IBD.[8] The number of metabolites is down-regulated in patients with IBD, featured by long-chain fatty acids, phenylbenzodioxanes, and cholesterols. Only a few of them are both enriched in patients with UC and CD including sphingolipids, carboximidic acids, and bile acids.[9]

Short-chain fatty acids (SCFAs), especially acetate, propionate, and butyrate which make up more than 90% of SCFAs in the gut, play a vital role in maintaining energy homeostasis and regulating the mucosal immune response.[10] Alterations of SCFAs and associated SCFA-producing bacteria have been identified in patients with IBD; however, the results of these studies are inconsistent. A recent meta-analysis[11] based on case-control studies revealed that the reduction of SCFAs correlates with disease activity and classification. *Faecalibacterium prausnitzii*, a butyrate-producer and predominant fecal microbiota, was found to markedly decrease in patients with CD.[12] Similarly, the reduction of this butyrate producer has also been identified in patients with UC with an inverse correlation with disease activity.[13] However, the quantity of *F. prausnitzii* does not match the concentration of butyrate separated from the fecal samples at a single time point in cross-sectional studies.[14] To date, appropriate models are lacking for the evaluation of intestinal micro-environment dysbiosis, including structure shift and functional alteration of the microbiome. In addition, high data heterogeneity remains among different research groups, due to relatively small sample sizes and confounding factors such as diet, medication, and severity of colitis.[15-17] Moreover, the discrepancy in metabolite categorization and methods of data presentation made it hard to integrate different findings.

Mycobiome and its Inter-Play With the Microbiome

To date, marked progress has been made in illustrating the role of the bacterial microbiome in the pathogenesis of IBD; however, the fungal microbiome, so-called mycobiota, is often ignored. Variation in bacterial microbiota structure can be identified by 16S rRNA sequencing; however, only genus level of bacteria can be distinguished and no mycobiota information can be acquired. Analysis of mycobiota configuration and function through meta-genomics data is relatively arduous. On one hand, the absolute amount of fungi comprises only less than 1% of total micro-organisms in the intestine[18]; thus higher resolution of sequencing is needed for mycobiome identification. On the other hand, the database on available mycobiome is far from complete. Only five types of fungi have been identified in the fecal samples of pediatric patients with CD through meta-genomics.[19] In a recent study, 18S rRNA and internal transcribed spacer (ITS) sequencing targeting fungi were conducted in mycobiome studies, and a distinct fungal microbiome configuration was identified in patients with IBD.[20] Several mycobiomes including *Blastocystis* and *Saccharomyces* are correlated with microbial richness and diversity, and the prevalence of these mycobiomes is reduced in patients with IBD at the genus level.[21] However, there are no significant differences in mucosal-associated fungal microbiota between patients with IBD and healthy controls based on ITS2 sequencing.[22] Limited evidence has revealed a local increase of *Basidiomycota* in colitis-

associated carcinoma mucosa. In dextran sodium sulfate (DSS)-induced mice model, dysbiosis of fungal microbiota can also be observed.[23] However, it remains unclear how fungal microbiota directly or indirectly mediates colitis.

Deficiencies in fungal recognition pathways play a role in the pathogenesis of IBD. The host immune system recognizes fungi partially through a C-type lectin receptors-dependent mechanism, and intracellular signals activated by dectin-1/caspase recruitment domain-containing protein 9 (CARD9) promote immune responses to fungi.[24] Commensal gut fungi induce inflammasome activation and interleukin (IL)-18 maturation via CARD9 pathways, thereby reducing colitis severity and colitis-associated carcinoma. Moreover, a sharply altered gut fungal microbiota landscape, featured by an overload of resident gut fungi and predisposition to colitis, have been identified in CARD9$^{-/-}$ mice[25]; nevertheless, the trend in bacterial microbiota changes is less prominent. CARD9 modulates pathogen-induced colitis by controlling its virulence in gut microbiota-dependent mechanisms, and the genetic deficiency of CARD9 can be restored by polysaccharides diets, partially through depriving pathogens of an energy source.[26] Moreover, impaired tryptophan metabolism activity is observed in CARD9$^{-/-}$ mice, which is in accordance with the decrease of *Lactobacillus reuteri* and *Allobaculum* sp., whose capability of tryptophan metabolism is identified in culture-dependent methods.[27] Further research has revealed exogenous tryptophan metabolites can ameliorate the severity of colitis in DSS-induced CARD9-deficient mice. This seems to be a promising way to reshape the composition of fungal microbiota by targeting CARD9 pathways and its associated down-stream metabolisms under the tenuous equilibrium in colitis.

The inter-play of bacterial and fungal microbiota under chronic colitis has been frontier research. Notably, there has been a trend of concomitant alterations of bacterial and fungal microbiota in patients with IBD. Indeed, cross-talk between gut microbiota and mycobiota is crucial for the intestinal homeostasis. Fungal microbiota can counterbalance deterioration in bacterial composition changes and maintain intestinal homeostasis, especially in the acute colitis phase.[21] Fungal depletion by fluconazole up-regulates acute colitis in DSS-induced mice model. However, in a bacterial depletion model treated by ampicillin, vancomycin, neomycin sulfate, and metronidazole cocktail, DSS-induced colitis is down-regulated. However, in chronic recurrent colitis, fungal microbiota is not a protective factor and can harmfully translocate into the spleen and mesenteric lymph node, resulting in persistent colitis. There is a positive correlation among *Candida tropicalis*, *Serratia marcescens*, and *Escherichia coli* in the fecal microbiota of patients with CD, and *in vitro* biofilms have revealed that these organisms co-operate to form an anti-microbial agent-resistant environment. Infection of *E. coli* can facilitate the adhesion of *C. albicans* to the gut mucosa, thus aggravating intestinal inflammation.[28] The inter-play of bacteria and fungi mediates the severity of colitis. In the DSS-induced colitis mice model, the colistin-resistant *E. coli* strain strengthens the effect of *Saccharmomyces boulardii* and *C. albicans* on colitis inflammation amelioration and deterioration, which may be associated with its facilitation in the fitness of fungal colonization.[29] The inter-action of microbiota also depends on their metabolites to some extent. *C. albicans*, as a common colonizer, produces the quorum-sensing molecule, farnesol, which plays a vital role in biofilm formation and can facilitate the virulence transition of *S. aureus* through the acquisition of the antibiotic-resistant phenotype.[30,31] However, as a ligand of the farnesoid X receptor, this molecule can also act as an immune modulator in the mucosal immune response[32] and serve as a proliferation inhibitor in cancer stem cells,[33] producing a rather sophisticated effect in intestinal micro-ecology. Together, these findings throw light on the potential mechanisms of multi-organisms in the gut micro-environment as a whole.

The Emerging Field: Dynamic Alteration of Multi-Omics

Previous works, mainly focused on isolated "omics," failed to decipher the initiating trigger of IBD from a concept of holism, as this elusive disease was perplexed by multiple pathogenic factors. Recently, multi-omics approaches have garnered greater attention. This system biology methods, integrate different dimensional of microbiome functionality, involving intestinal microbiota composition, genetic potentials, and transcriptional activity of microbial ecology, and finally metabolic molecules, thereby revealing the holistic landscape of this sophisticated network in the pathogenesis of IBD. However, most of the multi-omics studies have been limited by small sample sizes and the restricted number of omics included.

Multi-omics analysis has been primarily used in the development of effective biomarkers for colitis prediction and classification. Multivariate models based on transcriptomics and metabolomics data from the biopsy samples of 58 individuals (active UC, quiescent UC, or non-IBD controls) have provided candidate biomarker panels for disease severity evaluation of UC with high accuracy.[34] Moreover, a self-learning model from a recent study identified several microbial and host features from a meta-genomics and meta-taxonomics data set of 40 intestinal biopsy samples (from individuals with CD or without IBD as controls), and these features obtained by in-depth multi-omics data processing predicted disease severity and treatment response in pediatric patients with CD.[35] These multi-omics analysis strategies are relatively easy to understand and do not involve the inter-relationship of different omics and functional cluster processing.

Notably, another type of multi-omics research, implementing parallel and integrative analysis of meta-transcriptomics and proteomics from a data set of 47 intestinal biopsy samples (paired inflamed and non-inflamed mucosa from individuals with CD and UC or non-IBD controls), revealed that nearly 20% of biologic processes including ribonucleoprotein biogenesis, assembly and mRNA processing have discrepancies at the RNA and protein levels; post-transcriptional and post-translational regulations might account for these weakly correlated processes.[36] With similar data generation and clustering strategies, a

cross-sectional study, applying meta-genomics and proteomics data of stool samples from six twin pairs with CD, identified considerable protein diversity that could not be traced by current reference genomic sequencing.[37] Translating the multi-omics data into the clinical application of precision medicine is the ultimate goal of intestinal micro-environment research. In a recent study, researchers identified impaired mitochondrial hydrogen sulfide detoxification functions and up-regulated hydrogen sulfide (H_2S)- producing bacteria in patients with CD through system biology approaches combining meta-genomics and proteomics data sets. One-quarter of microbiota, positively correlated with the severity of CD, can metabolize sulfur-containing amino acid into H_2S, among which *Atopobium parvulum* is the central hub of this network.[38] Further research has elucidated the colitogenic capability of *A. parvulum* in colitis-susceptible IL-$10^{-/-}$ model mice, and this effect could be alleviated by the H_2S scavenger bismuth or removal of commensal bacteria. These parallel and complementary omics analyses have converged genetic potentials and real metabolic activity together, pointing toward new targets and latent mechanisms for further investigation.

Several multi-omics studies have investigated the inter-play of host genetic factors and the intestinal microbiome. In a recent large-scale cross-sectional research, meta-taxonomics and single-nucleotide polymorphisms were used to further elucidate host-microbe cross-talk in patients with IBD.[39] Healthy individuals carrying IBD genetic susceptibility variants, including nucleotide-binding oligomerization domain-containing protein 2, autophagy-related 16 like 1, CARD9, which are associated with bacterial handling, have a declining trend of *Roseburia* spp. in abundance. A decrease of *Roseburia* spp., the butyrate-producing bacteria that participate in intestinal inflammation prevention and amelioration through regulatory T-cell regulation, has already been proven to be associated with the microbiota alterations in patients with IBD. Another multi-omics study, which included more than 10,000 patients with IBD and healthy controls, found that a mis-sense mutation of solute carrier family 39 Member 8 (SLC39A8) was associated with CD.[40] In addition, the mis-sense variant of SLC39A8, which is involved in macrophage stimulation, differentiation, and bacteria clearance through autophagy, was found to be relevant to specific microbiota compositional shift in healthy controls, strongly correlating with the bacteria perturbation in patients with CD. These large-scale studies have identified genetic risk factors through risk variant screening in patients with IBD, followed by microbiota composition correlation analysis in healthy controls and further verification in other cohorts, shedding light on the potential targets of host-microbiota inter-play in the pathogenesis of IBD.

To date, only a few studies have attempted to profile the gut ecosystem shifts via comprehensive analysis based on more than three omics. A large-scale longitudinal study applied corresponding meta-genomics, meta-transcriptomics, viromics, proteomics, and untargeted metabolomics during a 1-year follow-up of patients with IBD.[6] Overall, although different functional profiles obtained from paired meta-genomics, meta-transcriptomics and proteomics have been tightly coupled, the correlation between transcripts and protein is relatively weak, partially due to individual divergence of the host-microbiome inter-play and post-transcriptional modification. When it comes to inter-individual microbial composition discrepancy, the genetic influence is more pronounced than disease severity, phenotype, and antibiotics interventions. The most surprising observation is that the profiles of each omics have a periodic shift in non-IBD controls, whereas there are no extra excursions in patients with IBD during the disease course. Moreover, questionnaires recording diet only contributed a much smaller part to the intra-individual, intermittent microbiome shift than expected; however, the effect of diet is still of significance. The rapid and significant microbial variation often occurred within 2 weeks in the natural courses, and there should be an aggressive sampling approach in future longitudinal studies. However, to date, multi-omics approaches have not exploited their full potentials regarding original data, and a tremendous amount of information is virtually in hibernation because of omics processing and integration. It is still challenging to integrate different omics as a whole, not only because of the divergent types of data set in different multi-omics matrices that convey multi-dimensional molecular information, but also because the parameters that influence the association of each omics data may outnumber the sample size, further hindering correlation analysis.[41] Better and standardized methods are required for specimen sampling and adjustment of confounding factors, and more applicable integration strategies and more promising artificial intelligence should be introduced for massive multi-omics data handling and processing.

Application of Microbiome Modulation in IBD

Numerous evidence highlights that the intestinal microbiome is an important trigger in the pathogenesis of IBD. Microbial alteration in active colitis is characterized by the reduction of beneficial bacteria and overgrowth of pathogenic ones, which serves as the rationale of microbiome modulation through diet, antibiotics, probiotics, and fecal microbial transplantation (FMT) as a promising approach for the treatment of IBD.

Supplementary probiotics can lead to the induction, maintenance of remission, and colitis-associated carcinoma chemoprevention, targeting microbiome modulation. Probiotics are living micro-organisms that exert beneficial effects when they accumulate in adequate amounts in the host, probably through their activity in intestinal barrier maintenance, mucosa immunity mediation, and micro-environment regulation. It has been speculated that probiotic cocktails can have a synergistic effect in inflammation inhibition since the formulation contains different microbial strains that participate in immunoregulation.[42]

A recent study based on a Chinese population identified an adjuvant effect of a probiotic cocktail, Bifico, which is a mixture of *Bifidobacterium longum*, *Lactobacillus acidophilus*, and *Enterococcus faecalis*, in the remission

induction of both mild to moderate CD and UC in the combination of mesalazine. Compared with mesalazine mono-therapy, Bifico provides more effects on the down-regulation of pro-inflammatory responses, as reflected by serum level of C-reactive protein and IL-6. Long-term outcomes, mainly evaluated by clinical manifestations during a 1-year follow-up, have demonstrated a significantly higher recurrent rate of 32% for the standard mesalazine therapy compared with only 5% for those who received adjuvant Bifico.[43] To date, there has been no validated evidence on the chemo-preventive impacts of Bifico on the carcinogenesis of IBD. Recent studies have revealed that Bifico can exert inhibitory effects on colitis inflammation in DSS-induced mice model and can ameliorate tumor load in azoxymethane (AOM)/DSS-induced mice with colitis-associated carcinoma. Bifico pre-treatment can change the landscape of mucosa-associated microbiota, especially featured by the tremendous expansion of genus *Lactobacillus* and the reduction of several bacteria with malignant potential. In addition, this microbiota alteration correlates with the down-regulation of CXC chemokines secretion, the bioactivity of which has been reported in intestinal inflammation and colorectal cancer.[44]

VSL#3 is another probiotic cocktail that has been widely used in the remission therapy of patients with IBD. This probiotic mixture contains four species of the *Lactobacillus* genus, three species of the *Bifidobacterium* genus, and *Streptococcus thermophiles*. An early large-scale cohort study that randomized 90 patients with acute mild to moderate UC to adjunctive VSL#3 therapy or SASP, 5-ASA mono-therapy for 8 weeks found that patients in the probiotics co-therapy group had an earlier remission response and had better efficacy in symptoms, endoscopic, and histologic evaluation.[45] Another Italian multi-center, double-blind randomized controlled trial (RCT) led by the same group that randomized 144 patients with relapsing mild to moderate UC to VSL#3 or placebo treatment for 8 weeks without adjustment of standard concomitant therapy, found that VSL#3 was marginally effective on the induction of remission compared with placebo. However, this probiotic cocktail was significantly superior in improving the UC disease activity index (UCDAI), with an extra 20% of patients experiencing greater than 50% amelioration of UCDAI during the 8-week observation.[46] A similar effect of VSL#3 on the induction of remission of UC was obtained in an Indian cohort involving 147 randomized mild to moderate patients with UC, notwithstanding the total remission rate of VSL#3 was merely 40%.[47] However, the effect of VSL#3 on the maintenance of remission has not been fully validated in a large population. Only a small study enrolling 29 pediatric active patients with UC revealed a better efficacy of VSL#3 on the maintenance of remission.[48] There are still controversies about the value of probiotics for the adjuvant therapy of CD. A double-blinded RCT, enrolling 23 patients with CD without fistulas or isolated terminal ileum lesions, revealed an even higher relapse rate in adjuvant VSL#3 group during a 1-year follow-up.[49] By contrast, a recent study suggested a favorable effect for VSL#3 in maintaining remission of CD after ileocolonic resection. Although endoscopic recurrence rate in the first 90 days was only marginally lower among patients receiving VSL#3, patients who received VSL#3 for the entire 365 days had statistically lower mucosal inflammatory cytokines levels and endoscopic relapse rate than those who did not receive VSL#3 during the first 90 days after surgery resection, suggesting the value of early probiotics intervention in maintaining remission of CD after surgery.[50] The anti-carcinogenic properties of VSL#3 have been identified in the AOM/DSS-induced mice model, probably through the therapeutic effect on the inflammatory response, thus inhibiting the inflammation-induced acceleration of the process of carcinogenesis.[51] Interestingly, the supplementary of VSL#3 did not significantly alter fecal microbiota abundance of *Lactobacillus* or *Bifidobacterium* at the genus level. However, increased mucosa colonized *Bifidobacterium* and rebalanced mucosal bacterial abundance were identified after probiotic mixture supplementary.[52] However, only a few of these studies have described the same study design, probiotics supplementary regimen, and similar clinical phenotypes enrolled, making it hard to integrate these results.

Although there is no doubt that microbiota regulation through probiotics supplementary can be a promising therapeutic target for IBD, the inconsistent therapeutic response in patients with distinct clinical entities suggest a sophisticated strategy to distinguish patients with proper phenotypes who may benefit from supplementary probiotics.

FMT is another strategy that directly manipulates microbiota in patients with IBD [Figure 2]. The idea of FMT may have originated from ancient Indian medicine. It was introduced to China in accompanied by Buddhism during the Han dynasty, and the clinical practice of FMT was first recorded in a traditional Chinese medicine handbook of emergency medicine (*Zhou Hou Bei Ji Fang*) centuries later, mainly for the treatment of acute diarrhea and food poisoning, the longer fermentation time of feces, the better efficacy that oral consumption provides.

The efficacy of the application of FMT on the induction of remission of IBD has been demonstrated in several studies, with an average clinical remission rate of 52% and 33% in CD and UC separately at the first treatment response evaluation, 2 weeks to 2 months after FMT.[53] However, most of these studies were non-controlled trials with divergent research designs and backgrounds. Patients adopted donor-like microbial configuration to some extent after FMT, and responders tend to have more obvious microbiota shift.[54,55] Although responders and non-responders share similar microbiota diversity, several specific genera might be promising predictors in response evaluation. Responders were identified to have a higher abundance of *Fusobacterium* and Enterobacteriaceae levels at baseline before FMT.[55] Besides, remission was also associated with the presence of several metabolites signatures, such as SCFAs and secondary bile acids.[56] Moreover, patients with marked alterations of *Collinsella* and Lachnospiraceae during microbiota supplement tended to have better treatment efficacy.

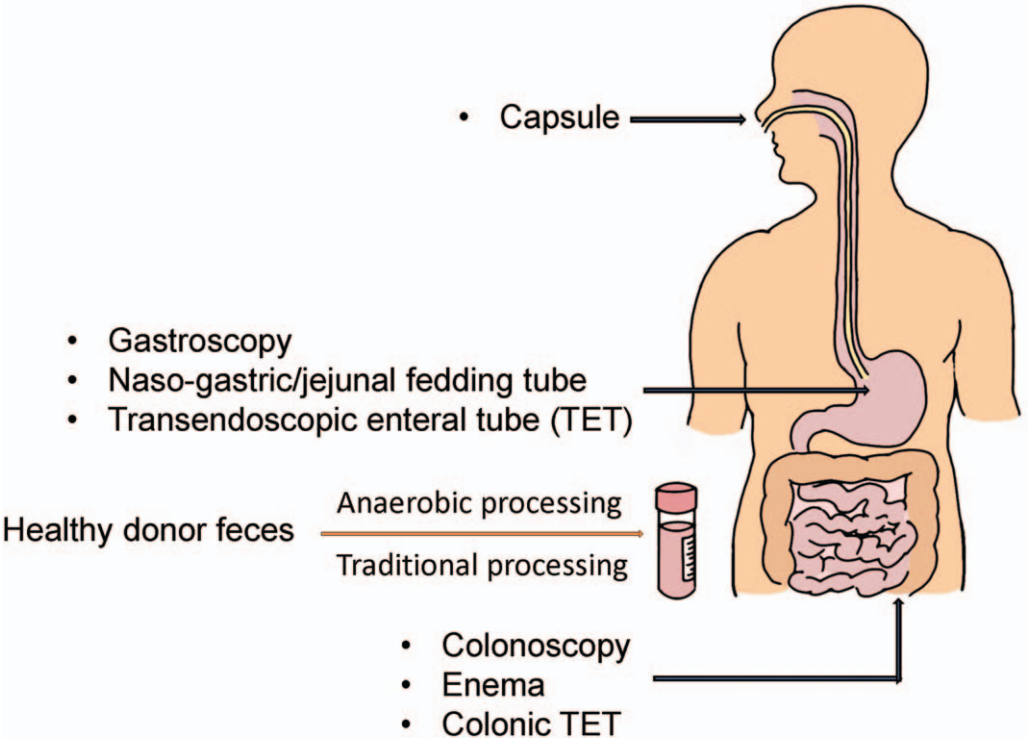

Figure 2: Overview of the common fecal microbiota transplantation regimen.

In previous studies, most of these clinical remissions or responses after FMT have been achieved in combination therapy with steroid or immunosuppressive agents, and the colitis still relapses during a longer follow-up period. To determine the optimal strategies for a second FMT to maintain a long-term remission, a retrospective study based on 69 patients who responded to the first FMT without combined steroids or immunomodulators identified a nearly 4-month median maintenance time of clinical response to the first FMT. Moreover, later serial FMTs could extend the progression-free intervals and prevent treatment failure.[57]

More recently, studies have focused on the efficacy of FMT on patients with specific phenotypes. Data from an Indian prospective study revealed that FMT via colonoscopy every 4 weeks could assist patients with steroid-dependent active UC to get rid of steroids successfully and ameliorate mucosal damage. At the end of this trial, 93.9% of patients who accomplished the seven rounds of FMT as per protocol withdrew steroids completely, 78.8% of patients obtained endoscopic remission in the intention-to-treat analysis.[58] Moreover, another large scale double-blinded RCT, which compared FMT and sham procedure with strict anaerobic stool processing methods for the steroids free remission of UC, indicated a resolution of disease without steroids observed in 32% and 9% patients, respectively, at 8 weeks. Aside from the mediocre steroid-free response rate, nearly 47% of patients who received three doses (one colonoscopy and two further enemas within 7 days) of the FMT regime achieved clinical remission, which was far better than the average level published in previous uncontrolled trials.[59] The relatively convenient short term, low-intensity FMT regime, and anaerobic samples processing which restored virtually all landscape of donor microbial composition, could be a promising strategy for further validation.

Dozens of questions remain about FMT, especially the efficacy of different routes of microbiota transplantation. It has been speculated that FMT through the upper gastrointestinal tract may not be as effective as the trans-anal route because the high acidity in the gastric cavity could destroy much of the microbial component. A recent RCT, recruited 27 patients with mild to moderate active CD, attempted to unearth the facts. Fresh donor feces obtained on the morning were transplanted to patients via gastroscopy or colonoscopy after sample preparation once a week for 2 weeks, an extra routine dose of proton pump inhibitor was provided on the evening before and the morning of FMT in the gastroscopy group to maintain active and living organisms as much as possible. The poor microbiota diversity in patients with CD resumed after microbiota transplantation in both groups as expected. Besides, it was intriguing to see the two strategies of FMT exhibited no significant differences in clinical remission and the main adverse events rate during the 8 weeks follow-up.[60]

FMT through oral capsules was another optional dosage regimen, which could be well accepted because of the convenience and sustainable discomfort. However, there are very few pilot studies assessing the efficacy of oral capsulated FMT in the remission therapy of patients with IBD. Preliminary evaluation from two small-sized non-controlled trials revealed dilatory improvement was achieved in the adjuvant oral application of encapsulated microbiota.[61,62] Despite the inconsistent alteration trend

of microbial diversity in different studies, patients who accepted FMT by oral capsules following broad-spectrum antibiotics establish a microbiota composition with similar diversity to donor prior to clinical response. In general, there is still a long way to go, especially the standardization of pre-treatment strategies and specific controlled release formulations of capsules on targeted patients, before a firm conclusion achieved.

Summary

IBD emerges through a rather complicated inter-action network of host genetics, environmental influencing factors, gut microbiome, and intestinal immune responses. Therefore, the practical value of isolated microbiota composition and diversity differences in cross-sectional designed research might be limited, prospective longitudinal trials illustrating dynamic alteration of multi-omics that reflects the extrinsic and intrinsic trigger could become a more promising strategy. Future efforts to IBD associated intestinal micro-environment shifts should pay more attention to the standardization of environmental confounding factors, like dietary and oral medications. Moreover, the functional community identified in multi-omics research should be further verified in germ-free mouse models to delineate the potential mechanisms and ultimately applied to the transplantation of specific micro-environment to patients with corresponding phenotypes.

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Conflicts of interest

None.

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