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Current and future targets for faecal microbiota transplantation

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ABSTRACT

The human gastrointestinal tract is home to the most diverse microbial ecosystem in the human body and is made up of bacteria, viruses and eukarya. Collectively known as the gut microbiota, our knowledge of these microbial communities has historically been restricted by the relative limitations of culturing techniques. However, the recent development and utilisation of next-generation sequencing techniques has enhanced our understanding of its structure, diversity and function.

There is emerging evidence that the gut microbiota plays a pivotal role in both health and disease. Perturbations to the structure and function of the gut microbiota are known to be associated with certain disease states. Therefore, manipulating the gut microbiota in an attempt to restore structure and function represents a promising therapeutic strategy. Recently, there has been a surge in clinical and scientific interest in manipulating the gut microbiota using a method called faecal microbiota transplantation. This increase in interest has gathered after it was shown in randomised controlled trials to be highly effective in treating recurrent *Clostridium difficile* infection.

Despite success in treating recurrent *Clostridium difficile,* there remain many unknowns about how best to optimise its preparation, regulation, mode of delivery and safety. This review aims to summarise the literature surrounding the current knowledge regarding faecal microbiota transplantation and explore potential future research avenues that aim to enhance the safety, efficacy and utilisation of faecal microbiota transplantation.

1. Introduction

The human gastrointestinal tract is home to the most dense, rich and diverse microbial ecosystem in the human body. The highest concentration of microbes live within the colon, where there are an estimated 10^{12} cells per gram of intestine luminal contents [1]. These microbial communities, the habitat they live in, and their spectrum of activity are collectively known as the gut microbiome. In this review, the term 'microbiota' will be used when reference is made to only the microorganisms themselves and the term 'microbiome' will be used when referring to the microbial communities, their genetic potential and the environment that they occupy.

The gut microbiome is composed of archaea, bacteria, viruses and eukarya [2]. The vast majority of research into the gut microbiome that has been conducted to date has focussed on the structure and function of the bacterial communities. By contrast, there has been relatively sparse research conducted on the viruses and bacteriophages (virome) [3], fungi (mycome) [4,5] and other micro-eukaryotes such as protozoa [6]. For the purposes of this review, the term 'gut microbiota' will refer to the bacterial communities that reside inside the intestinal tract, unless stated otherwise.

Until relatively recently, the majority of knowledge relating to the gut microbiota has been acquired through culture-based techniques, which are labour-intensive and not high-throughput. Furthermore, they require specific conditions to optimise bacterial growth (e.g. an anaerobic environment) which inevitably means that much of the gut microbiota is missed. The invention and subsequent implementation of next-generation sequencing technologies have provided researchers

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with the apparatus and capabilities to analyse the gut microbiota without the need to culture microbes [7]. Several international studies and initiatives, including large-scale endeavours such as the Human Microbiome Project and MetaHit, have used these tools to identify over 1000 species within the gut, mainly belonging to four major phyla, namely: *Actinobacteria, Proteobacteria, Firmicutes,* and *Bacteroides* [2,8]. These large cohort studies also showed that relative abundance of these phyla differs between individuals [9]. However, the clinical and biological significance of these observations is currently poorly understood.

Microbial sequencing enables us to know which microbiota community members are present but are unable to elicit their specific role. In view of this, numerous experiments have been undertaken to better understand the function of the gut microbiota [10,11]. These experiments have shown that the bacterial communities underpin several important physiological functions such as nutrient absorption, bile and short chain fatty acid metabolism, activity of the immune system, vitamin production and protection from xenobionts [12]. The functions of the gut microbiota appear to be ubiquitous across the healthy population [9]. In contrast, the structure and composition of the gut microbiota appears to differ between people. The significance of differences in structure and composition of the microbiota between person to person is currently unclear. However, it is known that a sensitive and complicated symbiosis exists between humans and their microbial inhabitants. Disturbances to this symbiotic relationship is known to have deleterious effects on the host. For example, in the case of *Clostridioides* (formerly *Clostridium*) *difficile* infection (CDI), the use of broad-spectrum anti-microbial agents disrupts normal microbial diversity and function. This in turn, allows germination, colonisation and toxin production by *C. difficile*, resulting in clinical symptoms of diarrhoea often associated with blood, pain and significant morbidity and mortality if left untreated. Therapeutic modalities focussed on restoring gut microbiota diversity and function are emerging and represent a promising therapeutic strategy. For example, a medical treatment called Faecal Microbiota Transplantation (FMT), has shown efficacy in randomised controlled trials for treatment of recurrent CDI [13].

This review provides an update on current understanding of FMT for CDI and gives an insight into best practice for all aspects of the treatment. The evidence for FMT in indications beyond CDI is also discussed.

2. *Clostridioides difficile* infection (CDI)

Clostridioides difficile, is an anaerobic, spore-forming, toxin producing bacterium that colonises the intestinal tract of around 2–5% of the healthy population [14]. It was first isolated by Hall & O'Toole in 1935 from the gut of a healthy newborn baby [15]. However, its implication as a human pathogen was not known until 1978, when George and colleagues discovered that *Clostridioides difficile* was heavily implicated in many cases of antibiotic-associated diarrhoea. These days, it is known as the causative pathobiont in most cases of post-antibiotic infectious diarrhoea [12]. In 2011, there were approximately 500,00 cases of CDI, resulting in 29,000 deaths in the USA [16]. The European Centre for Disease Prevention and Control estimated using a point-prevalence survey that ~124,000 patients developed health-care-associated CDI within the European Union annually.

A detailed description of the pathogenesis of CDI is beyond the scope of this review. The major risk factor for the development of CDI is the use of broad-spectrum antimicrobial agents [17], which has led many to hypothesise that in health, the indigenous gut microbiota functions to prevent the germination and overgrowth of *C. difficile* (as well as other enteropathogens), a concept known as 'colonisation resistance'. Potential mechanisms driving this phenomenon include the production of bacteriocins and phage by the indigenous communities, and alterations of gut microbiota-host metabolism interactions – which may impact competition for nutrients and physical space [18,19]. As one example of interest, restoration of gut microbiota-mediated bile acid metabolism has been demonstrated after successful FMT for rCDI [20]. Particular bile acids have been demonstrated *in vitro* to have profound effects on different parts of the *C. difficile* life cycle, with the primary bile acid taurocholic acid being a major progerminant, and secondary bile acids (e.g. deoxycholic acid) inhibiting *C. difficile*'s vegetative growth [21]. As such, the degradation of taurocholic acid and enrichment of gut secondary bile acids that accompanies FMT for rCDI [20] may be an important mechanistic explanation for FMT's success.

There are a broad range of clinical manifestations of CDI, ranging from mild diarrhoea to life-threatening toxic megacolon [22]. First line treatment for CDI involves stopping the inciting antibiotic and rehydrating the patient. Following this, the recommended treatment is a course of antibiotics. Those currently licensed in the EU are: metronidazole, vancomycin and fidaxomicin. Initial treatment is generally successful [23], however, the risk of recurrence within eight weeks is 15–25%, which rises to 40–65% in patients that have had more than a single recurrence [24]. In patients with multiple relapses, the treatment options include: a pulsed/ tapered course of vancomycin or fidaxomicin, a novel anti-toxin B monoclonal antibody called Bezlotuzumab [25,26], and faecal microbiota transplantation (FMT). Despite these treatment options, there has been the emergence of hypertoxigenic strains of CDI, including 027, which are less responsive to antimicrobial treatments and therefore drive the need for alternative treatment options [27].

3. Faecal microbiota transplantation (FMT): history and definitions

In the late 1950's, the Chief of Surgery at Denver General Hospital, Mr Ben Eiseman, decided that he would try to treat four of his patients suffering from post-antibiotic diarrhoea by transferring stool from a healthy donor into their intestinal tracts with good results. Since the pioneering work of Eiseman, a large body of controlled and non-controlled evidence has accumulated showing that FMT is a highly effective therapeutic strategy in patients suffering from recurrent CDI [28]. A detailed analysis of the literature is presented in further detail in a later section of this review.

A recent systematic review published by Quraishi and colleagues reported that across seven randomised trials and 30 case series, FMT was more effective than vancomycin in resolving recurrent and refractory CDI, with clinical resolution across all studies being 95% [28]. In contrast, the efficacy of vancomycin in patients with recurrent CDI not responsive to multiple courses of antibiotics is known to be around 30% [24]. In addition to recurrent and refractory CDI, a recent study reported that FMT also improves survival in patients suffering from severe CDI [29].

The majority of FMT reported in the literature is allogenic in nature (Fig. 1) and in volves the transfer of faecal microbiota from a healthy donor into the intestinal tract of a recipient. However, it should be noted that FMT can also be autologous in nature (Fig. 1), where faecal microbiota is banked by a person and reinstated at a later date, potentially after medical treatment that alters the structure and/or function of the gut microbiome. A variety of methods can be utilised to deliver the faecal microbiota as part of the procedure, such as nasoduodenal tube, nasogastric tube, rectal enema and the biopsy channel of a colonoscope [14]. Recently, there has been interest in delivering the faecal microbiota through enteric coated capsules [30,31]. Enteric coated capsules aid the design of placebo controlled studies and remove the need for the invasive medical procedures currently used to administer the faecal microbiota. However, in the studies, patients were asked to swallow 30–40 capsules, which could potentially pose problems for certain groups of patients in particular, e.g. with swallowing disorders. In light of this, the optimal delivery method remains unclear and future research should determine if the capsule burden can be decreased [32].

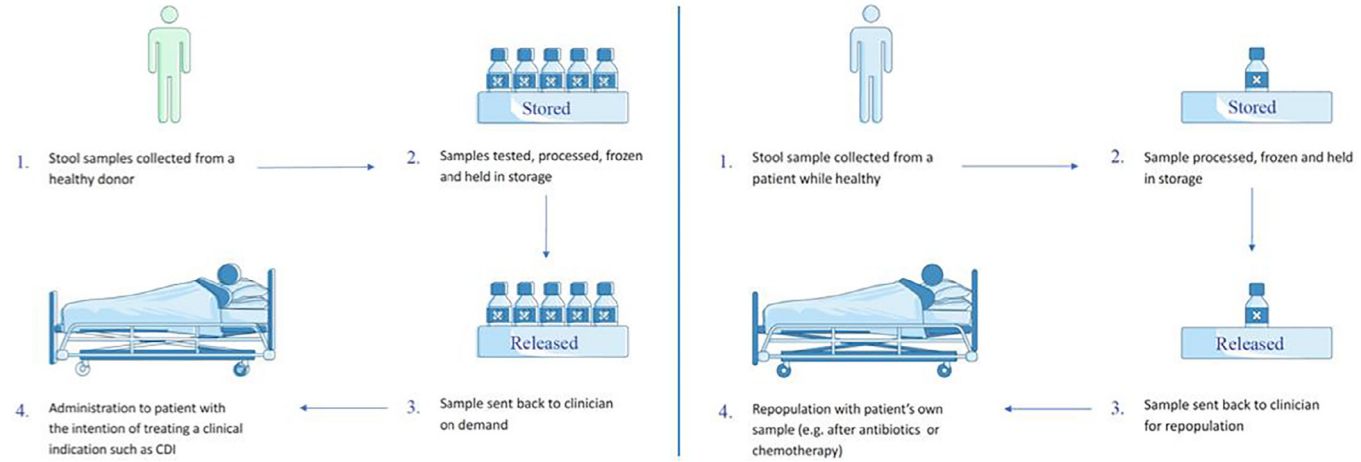

Fig. 1. A schematic overview of allogenic and autologous Faecal Microbiota Transplantation using banked frozen faecal microbiota.

4. Current regulatory landscape

There is worldwide variation in the regulation of faecal microbiota for therapeutic application [33]. In some countries, such as the UK [34], the USA [35] and France [36], faecal microbiota is regulated as a medicinal product. In others, such as Italy, it is regulated as a tissue and in others, such as the Netherlands, there are currently no regulatory guidelines [37]. In the USA, despite announcing that faecal microbiota is regulated as a medicinal product, the FDA has opted to exercise enforcement discretion for FMT used for recurrent or fulminant CDI which fail to respond to standard therapy [38]. In Europe, the EU commission provided a legal opinion on the regulation of FMT in December 2014. The commission considered that for the purposes of the EU Tissues and Cells Directive (EUTCD), faecal microbiota is a 'combined substance', meaning that it contain human cells and other components not from human origin. There is a precedence for combined substances falling under the EUTCD. However, the commission concluded that for the purposes of FMT, the cells are not the active component of this substance and therefore are not 'intended for human applications' within the definition of the EUTC. In the future, it is possible that these policies and stances will evolve in time as the evidence base for FMT matures and progresses.

5. FMT practicalities in clinical practice

5.1. Donor screening and selection

Historically, patients were encouraged to select their own donor from friends and family. However, evidence has emerged showing that faecal microbiota obtained from unrelated 'universal donors' is equally efficacious [30]. Furthermore, there is evidence from blood transfusion medicine showing that recipient-selected donors are not less likely to test positive for infectious disease than unrelated 'universal' volunteer donors [39]. These data, coupled with evidence showing that faecal microbiota can be frozen with a cryoprotectant and banked for over six months at −80 degrees Celsius [40] without a loss of viability and efficacy [13] has prompted many centres to set up donor programmes using pre-screened unrelated donors who are anonymous to the recipient. Frozen banking also allows for stool to be quarantined, which has the added benefit of reducing the risk of infectious pathogens not being picked up because of screening being performed during a seroconversion window. The concept of frozen banking is well established, however, the duration of quarantine and the frequency of retesting is less well agreed.

Currently, there is no consistent evidence that links donor characteristics to any influence on patient outcomes. In light of this, donor screening is focussed on risk reduction [41]. Consensus guidance published by Cammarota et al. [32] recommends that donors are extensively screened by a medical questionnaire prior to undergoing blood and stool testing. The medical questionnaire is usually designed to elicit information regarding risk factors for transmittable pathogens and conditions and diseases that could potentially be microbiome-mediated [32]. As a general rule, prospective donors with active infection or who disclose risk factors for infection should be excluded. Regarding microbiome-mediated conditions and diseases, as FMT involves the transfer of a largely uncharacterised active microbial suspension, there is a theoretical possibility that a propensity to diseases linked to the gut dwelling microbial communities could be transferred [6]. Therefore, it is recommended that centres and doctors adopt microbiome-specific exclusion criteria, such as diagnosed metabolic disorders and obesity, a personal or family history of gastrointestinal disease and a family history of colorectal cancer [42].

There is relative uniformity different centres and countries (Table 1) with regards to blood screening protocols. By analogy, it may be best practice to try and match the serostatus of donors to patients for CMV and EBV which is likely to be an important consideration in immunocompromised recipients.

In contrast to the general uniformity and convergence observed in Table 1 between donor blood testing protocols, stool screening protocols vary between centres and countries [37]. In general, clinical trials appear to adopt similar testing protocols to those outlined in the seminal randomised controlled trial published by Van Nood et al [13,44,45]. Between stool banks, most of the variation between protocols is found between policies on screening for multi-drug resistant

Table 1
A table outlining blood testing protocols in a number of commercial and non-commercial stool banks. These are presented alongside EU consensus guidance published by Cammarota et al [32].

	OpenBiome [43]	NDFB [37]	EnteroBiotix	EU Guidance [32]
Test				
CMV	–	X	X	X
EBV	–	X	X	X
Hepatitis A	X	X	X	X
Hepatitis B	X	X	X	X
Hepatitis C	X	X	X	X
Hepatitis E	X	X	X	X
HIV Type 1/2	X	X	X	X
HTLV	X	X	X	X
Treponema	X	X	X	X
Strongyloides	X	X	X	X

organisms, as well as contagious viruses such as Astrovirus and Sapiovirus [37]. In the absence of robust data on optimal donor testing and management, centres should take a multi-disciplinary approach to developing screening protocols and processes. The current consensus guidance criteria for screening and subsequent selection will likely be updated to reflect improved understanding of transferring potential pathogens through FMT.

5.2. Recipients of FMT

FMT works for recurrent CDI, and probably also refractory CDI, although refractory is a term that is poorly-defined. Interestingly, recent data suggests that FMT may be a suitable alternative to antibiotic treatment in primary CDI [46] although numbers in the studies are small to make definitive conclusions and recommendations. There are many unknowns about who should and shouldn't receive FMT. Many studies involving FMT have excluded patients who are immunocompromised, pregnant, [47,48] as well as those with chronic diarrhoea[47–49], decompensated cirrhosis [48] and those with food allergies [48,50]. Whilst there is little evidence that these exclusions are associated with significant risk from FMT, it should be offered with caution in these patient groups and based on theoretical risk avoided in those patients with anaphylactic food allergies.

5.3. Faecal processing and storage

The methods, techniques and processes for faecal collection and preparation prior to administration have not been standardised and vary across the literature. Guiding principles have, however, been published in consensus papers.[32,51] Faecal material should be collected from donors in a designated clean air tight collection container. Once the stool has been obtained, it is advisable to process the sample within six hours [49] to preserve as much viability as is practically possible. European consensus guidance recommends that > 30 g of faecal microbiota is used per FMT. However, we recommend that this is increased to > 50 g of in light of evidence showing that that using < 50 g increases the risk of CDI recurrence [52]. Once weighed out, stool is usually homogenised with an excipient so that it can be infused as part of the FMT procedure. The most commonly used in the literature is physiological saline, however, other excipients such as water [53] and milk [54] have been described previously. After homogenisation, the faecal microbiota is usually filtered to remove large particulates. If the material is to be frozen, then glycerol or another cryoprotectant should be added to the suspension to preserve the viability of the bacterial communities when the suspension is frozen. There is data showing that faecal microbiota remains viable and efficacious for six months when frozen with glycerol [45].

5.4. FMT preparation and procedure

On the day of the procedure, frozen faecal microbiota preparations should be thawed overnight in a refrigerator or at room temperature [55] for several hours. The Netherlands Donor Faeces Bank recommends 5 h of thawing if at room temperature [37]. Thawing the preparation in a water bath is not recommended as there are published reports of water baths causing microbial entry during the thawing process in blood transfusion medicine [56].

Patients undergoing FMT are typically asked to discontinue antibiotic therapy 1–3 days before the procedure. European consensus guidance also recommends the use of bowel lavage the day before FMT. It has been suggested that this may reduce the abundance of *C. difficile* and potentially remove any residual antimicrobial in the colon, and enhance engraftment of the donor faecal microbiota [51] however these hypotheses have yet to be tested in a robust randomised controlled clinical trial.

Further patient preparation to be considered is the use of proton pump inhibitors (PPI) prior to upper GI FMT. This theoretically helps to minimise acidity which may impair engraftment of transplanted microorganisms. Importantly, whilst some studies advocate the use of PPI prior to receiving FMT via the upper GI route [57–63], there appears to be no difference in efficacy when PPI has not been utilised. The use of a prokinetic for FMT via upper GI route has been described [64] and theoretically can reduce the risk of aspiration when utilising this route. When considering lower GI administration loperamide and other antimotility medications have been utilised with the aims of prolonging FMT exposure to the gut and aid retention of the FMT [65–67]. Whilst these additional patient preparation points should be considered, there is limited evidence for their use.

As briefly outlined earlier in this review, there are several potential routes of administration for FMT. These are: (a) instillation of faecal microbiota into the upper GI tract through nasogastric or nasoduodenal tube [13], (b) instillation into the colon by colonoscopy [44] or flexible sigmoidoscopy(c) instillation into the colon/rectum through an enema [53] and finally (d) administration through orally-delivered capsules [30]. The optimal route of delivery remains unclear, with each having potential benefits and drawbacks. For example, upper GI administration may require less sedation than colonoscopy but several studies have shown that colonic delivery may provide a slight, but not statistically significant, efficacy benefit [28,58,68]. In addition, there have been several documented cases of aspiration pneumonia [59,69] as a result of upper GI delivery (see Section 7). Further RCT's are required in order to establish the optimal route of administration. Clinicians seeking to administer FMT should approach each clinical situation where FMT is potentially indicated on a case by case basis, taking the patients comorbidities and preferences into account.

6. Fresh vs frozen

Two randomised studies have evaluated the efficacy of fresh FMT versus frozen FMT. In one non-inferiority study, it was found that that a frozen enema of FMT (n = 91) was non-inferior for clinical resolution of diarrhoea to fresh FMT (n = 87) for the treatment of recurrent or refractory CDI [55]. The other study supported this finding, and suggested that remission rates for CDI were similar when comparing fresh and frozen FMT delivered via colonoscopy (n = 25/25 vs 20/24 respectively, p = 0.233 [65]).

On a logistical level, frozen FMT is likely to be the preferred choice due to ease of transferring from centralised stool banks, traceability, and from a regulatory perspective. Research has shown that stool banking is more cost effective than using directed donors and fresh samples [70]. Furthermore, research has demonstrated that frozen stool retains its viability and efficacy for six months when stored in a −80 °C freezer [45,50,70], however decreasing viability of the gut microbiota has been shown beyond this period.

7. Adverse events and unintended consequences

Those adverse events reportedly occurring around the time of administration of the faecal microbiota FMT tend to be related to the procedure itself. Procedural adverse events described after colonoscopic administration include mild nausea and vomiting (attributed to sedation for the colonoscopy), and minor mucosal tears during colonoscopy. A case of microperforation [71,72] following biopsy of an area of possible small bowel ischaemia in a patient with chronically dilated small bowel has also been reported; the case was subsequently successfully treated conservatively[66]. One death due to witnessed aspiration at the time of colonoscopy has been described [73]. Two deaths related to aspiration pneumonitis that are likely attributable to upper GI administration of FMT have been reported [73,74] along with several cases of regurgitation and vomiting [74]. However, some of these patients were receiving a considerably higher volume of FMT than is typically administered by the upper GI route (i.e. 500 ml) [74]. One

of the fatal episodes using higher volumes occurred in a patient with a swallowing disorder following oropharyngeal radiation after surgical removal of a maxillary carcinoma. Many centres using smaller volumes of FMT for upper GI administration have not consistently noticed similar problems.

In the hours to days following FMT, the most common adverse events are constitutional or gastrointestinal symptoms, including diarrhoea, abdominal cramps/pain, belching, constipation and nausea [74]. These are typically mild and self-limiting. Successful treatment usually centres around conservative management. However, it should be noted that a recent *meta*-analysis described a 22.7% rate of worsening of IBD activity in patients with IBD that received FMT as treatment for rCDI [75] but the authors of this study did note significant heterogeneity in the reviewed literature. Further studies are required to further delineate the relationship between FMT and risk of IBD flare.

There are concerns about the risk of transmission of infection from donor to recipient through FMT. However, there are very few reports of this in the literature. Two cases of Norovirus infection occurring soon after FMT have been reported, however, the authors concluded that these infections were more likely to represent environmental transmission rather than direct donor-to-recipient transfer [76].

Beyond what has been described above, there have been reported cases of autoimmune and inflammatory conditions developing soon after FMT. These include: microscopic colitis, Sjögren's syndrome, follicular lymphoma, peripheral neuropathy, immune thrombocytopenia and rheumatoid arthritis [77,78]. It should be noted, however, that these observations have only been published in case reports and have not been replicated in randomised controlled trials. A widely-publicised case study reported marked weight gain in a patient being treated for recurrent CDI using an overweight family member as a donor [79]. These results have not been replicated elsewhere in the literature and therefore does not seem likely to be a true risk [80].

As faecal microbiota contains a largely uncharacterised consortia of microorganisms, there is a theoretical risk that FMT could transfer a disease phenotype from the donor to the recipient. Long-term, prospective follow-up of recipients is required to fully assess this risk and donor screening programmes should be designed to exclude donors that are most likely to harbour significant risk.

8. Efficacy in Non-CDI indications

The role of FMT has been explored in a multitude of indications beyond CDI. Most of the research that has been conducted has been uncontrolled and there is significant heterogeneity between case reports and case series that report positive outcomes [81]. There is likely to be a significant degree of reporting bias and therefore the positive outcomes should be approached with caution. However, there have been studies published in several indications beyond CDI, including: ulcerative colitis, metabolic syndrome, functional bowel disorders, hepatic encephalopathy and multi-drug resistant organisms.

8.1. Ulcerative colitis

To date, there have been four RCTs (three published, one abstract) investigating clinical endpoints in UC following treatment with FMT [44]. Three of these four RCTs showed positive outcomes, with patients receiving donor FMT more likely to reach clinical endpoints of response and remission compared to placebo or autologous FMT. However, these studies differed in methodology, with variations in patient inclusion criteria, donor stool processing and preparation, administration and the faecal microbiota infusions. The successful studies delivered the faecal microbiota through the lower GI route and used a more intense treatment protocol with up to a total of 40 over 8 weeks. Interestingly, the RCT that prepared the faecal microbiota under anaerobic conditions resulted in the highest clinical response and remission.

8.2. Functional bowel disorders

There have been two RCTs that have been published investigating the use of FMT as a treatment for functional bowel disorders. These two studies report positive outcomes. A Norwegian study of 90 patients with diarrhoea-predominant irritable bowel syndrome (IBS) demonstrated that a an improvement in IBS severity scores in those who received a single infusion of FMT compared to those who received placebo [82]. A second study showed marked improvement in spontaneous bowel movements in 60 patients with slow transit constipation receiving six infusions of FMT when compared to conventional treatment [83].

8.3. Metabolic syndrome

To date, there have been two studies investigating the efficacy of FMT in patients with metabolic syndrome [84]. Both of these studies suggested that FMT from lean donors may improve peripheral insulin sensitivity in patients suffering from metabolic syndrome. In both studies the effect size was deemed to be statistically significant. However, the effects were transient (with benefits found only at six weeks – but no longer – post-FMT), which suggests that long-term repeated dosing may be required if FMT is to potentially be an effective treatment option in metabolic syndrome.

8.4. Hepatic encephalopathy

In a study of 20 patients suffering from hepatic encephalopathy caused by liver cirrhosis, improvements in encephalopathy and a reduction in cirrhotic complications were noted in the FMT arm, but not amongst patients receiving standard of care medical therapy [85]. However, as the patients in the FMT arm in this study also received antibiotics (rifaximin) and lactulose throughout, the results should be interpreted with caution.

8.5. Decolonisation of multidrug resistant organisms

There is a growing interest that FMT may promote decolonisation of multidrug resistant organisms [86–88]. As yet there are no randomised controlled studies that explore this but it is gathering interest and may provide a novel therapeutic avenue to target multidrug resistant organisms.

9. Conclusion and future perspectives

FMT is an effective treatment for recurrent CDI regardless of the route of delivery and method of preparation and storage. The use of encapsulated and orally administered faecal microbiota will expand access for patients and simply the design of placebo controlled trials. Short-term follow-up suggests that FMT appears to be a relatively safe treatment, with the majority of side effects being mild and self-limiting. The long-term sequelae of FMT are currently not known and future work should focus on investigating this. There is emerging evidence suggesting that FMT may have a treatment utility beyond recurrent CDI, although further RCT evidence is required before wide scale adoption may occur for these indications. In the future, it is possible that FMT may ultimately be replaced by defined consortia of bacteria or single strains that have been rationally-selected based on their mechanism of action. There are several commercial and non-commercial organisations and groups working on this [89]. However, it should be noted that faecal microbiota is a highly complicated starting substance and those that wish to reverse engineer it will probably have to elucidate the ways in which the microbial communities within the samples interact with each other, as well as unravelling its mechanism of action. In light of this, it may be quite some time before an optimal combination of microbes is discovered and ultimately taken into the clinic. Until then, doctors and researchers should focus on making FMT as safe

and effective as possible by adhering to consensus guidance recommendations and supporting interventional FMT studies.

10. Competing interests

JM received salary and travel support from EnteroBiotix Limited during the course of this study, but EnteroBiotix did not directly contribute to this work.

11. Authorship

All authors have made substantial contributions to the conception and design of the study, the acquisition of data, the analysis and interpretation of data. All authors have contributed to drafting the article and revising it critically for important intellectual content. All authors approved the final approval of the version to be submitted.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.humic.2018.08.004.

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