CHEST

Clinical Practice and Infrastructure Review of Fecal Microbiota Transplantation for *Clostridium difficile* Infection

Brendan J. Kelly, MD, MSCE; and Pablo Tebas, MD

A substantial proportion of *Clostridium difficile* infection (CDI) cases recur after completion of antibiotic therapy, and antibiotic cure rates diminish with each recurrence of CDI. Fecal microbiota transplantation (FMT) is an effective therapy for recurrent FMT, which otherwise requires prolonged or indefinite antibiotic treatment. FMT is performed by introducing the fecal microbial community obtained from a healthy donor or pool of donors into the stomach, small intestine, or colon of a patient with CDI. Multiple clinical trials support the usefulness of FMT in treating recurrent CDI, and CDI treatment guidelines now include consideration of FMT at the third CDI recurrence. However, there remain challenges to incorporating FMT into clinical practice. First, methods of fecal bacterial community processing vary, as do methods of FMT administration. Second, the optimal dosing strategy and expected benefit of FMT for refractory CDI, particularly for severe and severe complicated cases, are uncertain. Third, the US Food and Drug Administration (FDA) considers FMT an investigational treatment. Fourth, insurance reimbursement for FMT usually falls short of FMT administration costs. In the setting of rising *C difficile* incidence and growing evidence for FMT efficacy, the demand for FMT has increased. However, uncertainty surrounding optimal FMT preparation and administration methods, FDA oversight, and insurance reimbursement presently limits the clinical practice of FMT.

CHEST 2018; 153(1):266-277

KEY WORDS: *Clostridium difficile* infection; fecal microbiota transplantation; microbiome

Clostridium difficile infection (CDI) is a clinical syndrome typically characterized by loose, frequent bowel movements and abdominal pain, which occur as a result of toxin production by colonic *C difficile*.[1-4] CDI occurs in patients colonized with toxigenic *C difficile* and in patients who have newly acquired *C difficile*. Estimates of the relative contributions of persistent colonization and new acquisition to incident CDI vary.[5,6] In both cases, the pathogenesis of CDI involves depletion of non-*C difficile* colonic microbiota, altered bile acid metabolism, germination of (resident or recently ingested) *C difficile* spores, expansion of a population of vegetative *C difficile*, toxin production, and colonic inflammation.[7-12]

ABBREVIATIONS: CDI = *Clostridium difficile* infection; EIA = enzyme immunoassay; FDA = Food and Drug Administration; FMT = fecal microbiota transplantation; GDH = glutamate dehydrogenase; IBD = inflammatory bowel disease; IBS = irritable bowel syndrome; IND = Investigational New Drug; MDRO = multidrug-resistant organism; NAAT = nucleic acid amplification test; R-CDI = recurrent *C difficile* infection; SC-CDI = severe complicated *C difficile* infection

AFFILIATIONS: From the Division of Infectious Diseases and Fecal Microbiota Transplantation Program, Department of Medicine, Perelman School of Medicine, University of Pennsylvania, Philadelphia, PA.

FUNDING/SUPPORT: B. J. K. was supported by the National Institutes of Health [Grant K23 AI121485] and the Centers for Disease Control and Prevention Prevention Epicenters [Grant U54 CK000485].

CORRESPONDENCE TO: Brendan J. Kelly, MD, MSCE, Division of Infectious Diseases, Department of Medicine, University of Pennsylvania, 3610 Hamilton Walk, Johnson Pavilion 502, Philadelphia, PA 19104; e-mail: brendank@mail.med.upenn.edu

Copyright © 2017 American College of Chest Physicians. Published by Elsevier Inc. All rights reserved.

DOI: https://doi.org/10.1016/j.chest.2017.09.002

The primary event in CDI pathogenesis appears to be depletion of non-*C difficile* colonic microbiota with a loss of colonization resistance and a concomitant increase in *C difficile* spore germination.[8,13-16] The risk factors known to correlate with risk for *C difficile* colonization and CDI—antibiotic exposure, gastric acid suppression (with proton pump inhibitors, in particular), and age—each plausibly contributes to this process.[17-20] Consistent with current understanding of CDI pathogenesis, delivery of colonic microbiota from a healthy donor, or fecal microbiota transplantation (FMT), long has been known to eliminate the symptoms of CDI.[21-26] Such treatment appears to succeed in relieving the symptoms of CDI in proportion to its success in reconstituting the bacterial community of the donor, though complete engraftment is not necessary, and microbes other than bacteria may play a role as well.[27-37]

The first-line treatment for CDI remains anti-*C difficile* antibiotics (metronidazole, oral vancomycin, or fidaxomicin).[2,3] Treatment of CDI with anti-*C difficile* antibiotics is typically successful in relieving CDI symptoms, but anti-*C difficile* antibiotics may suppress other colonic microbiota, and *C difficile* may persist for months as a spore. As many as 20% of CDI cases treated with anti-*C difficile* antibiotics recur, and the risk of recurrent CDI (R-CDI) increases with each subsequent CDI episode.[38,39] Enthusiasm for FMT has increased as CDI incidence has increased, and clinical trials consistently have demonstrated the efficacy of FMT in resolving R-CDI.[27,31,40-45] However, the clinical use of FMT remains difficult because of uncertainty surrounding optimal methods of fecal bacterial community processing and FMT administration, as well as requirements related to US Food and Drug Administration (FDA) oversight and insurance reimbursement. Reviews provide valuable insight into CDI pathogenesis,[7,15,46] setting up a frozen stool bank,[47,48] FMT administration methods,[49,50] and the details of FDA oversight of FMT.[51,52] Here we review current FMT clinical practice, paying particular attention to issues relevant to FMT administration for patients who are hospitalized and critically ill.

CDI Diagnosis and Eligibility for FMT

Accurate identification of CDI is difficult but essential to the successful use of FMT. Accurate identification of CDI demands (1) evaluation of risk factors for *C difficile* colonization and infection, as well as the consistency of the manifesting syndrome with the common clinical features of CDI; (2) consideration of alternative diagnoses; and (3) assessment of stool *C difficile* test results. The first and second steps are crucial given ongoing debate regarding the diagnostic usefulness of the nucleic acid amplification test (NAAT) for toxigenic *C difficile*. NAAT is a first-line test for CDI according to current US guidelines.[2,3] However, NAAT has come under scrutiny because some studies suggest that NAAT may be too sensitive and fail to discriminate CDI from *C difficile* colonization. Patients with positive NAAT but negative enzyme immunoassay (EIA) results for *C difficile* toxin have less *C difficile* bacterial load, fecal inflammation, and diarrhea and fewer CDI-related complications.[53-55] Given this evidence, published guidelines recommend that positive NAATs be validated with a more specific toxin A and B EIA (a two-step protocol).[56]

The pretest probability of CDI is higher if one or more risk factors for *C difficile* colonization and CDI are present. Key risk factors include exposure to antibiotics, exposure to gastric acid suppressants (proton pump inhibitors, in particular), and age.[17,38,39,57-65] Health-care exposure is another risk factor for CDI, but health-care exposure must be understood to extend beyond hospitalization. A 2015 survey of 15,461 CDI cases in the United States demonstrated that 65.8% of cases were associated with health-care exposure but that only 24.2% started during hospitalization.[38]

The typical clinical manifestation of CDI includes abdominal pain and cramping accompanied by frequent, watery stools.[1,66] Melena and hematochezia are rare.[2,67] Signs of a systemic inflammatory response to colitis, including fever and leukocytosis, may be present, and the presence of such symptoms defines severe complicated CDI (SC-CDI). Symptoms typically improve within a week of starting appropriate anti-CDI antibiotics, though dose escalation may be required to induce a response in some cases.[3] Postinfectious irritable bowel syndrome (IBS), which can occur in the post-CDI period even after successful CDI treatment and may include prominent lactose intolerance, sometimes interferes with the full resolution of GI symptoms.[68]

The diagnosis of CDI typically is based on (1) NAAT, (2) a two-step algorithm that combines EIA testing for glutamate dehydrogenase (GDH) and *C difficile* toxins A and B, or (3) a three-step algorithm that uses GDH and toxin EIA testing up front and NAAT if GDH and toxin results are discordant.[2-4,69,70] As discussed, the

diagnostic validity of positive NAAT results in the absence of positive toxin EIA results is under dispute.[53,54] In our experience, it is essential to consider alternative diagnoses if NAAT results are persistently positive but response to high doses of anti-CDI antibiotics is equivocal. Inflammatory bowel disease (IBD) is a risk factor for both *C difficile* colonization and CDI. Particularly when hematochezia is a prominent clinical feature, response to appropriate anti-CDI therapy is minimal, and diagnosis of CDI rests on NAAT results alone, IBD should be considered.[68]

R-CDI is defined as CDI that recurs after initially successful therapy. Ten percent to 20% of appropriately treated CDI cases recur within 8 weeks of treatment, and the recurrence rate increases with subsequent recurrences.[27,71] Recurrences may indicate relapse of the same *C difficile* strain responsible for the primary infection or reinfection in the setting of persistent perturbation of the colonic microbiota.[28,72,73] R-CDI is the best established indication for FMT, with efficacy > 80% now supported by multiple randomized controlled trials. Current CDI treatment guidelines recommend consideration of FMT after the third recurrence.[3,27,31,40-45,74]

FMT also may be considered for cases of refractory CDI or SC-CDI. Refractory CDI, or CDI unresponsive to high-dose oral vancomycin or fidaxomicin, often fits the criteria for SC-CDI (ie, associated with hypotension, fever, ileus, abdominal distention, mental status change, leukocytosis or leukopenia, elevated serum lactate levels, end-organ dysfunction, or requiring intensive care unit admission).[3] Randomized clinical trial data are lacking to guide FMT treatment for SC-CDI. Observational studies suggest a possible benefit from FMT, but there is debate as to the optimal FMT dosing regimen in this population; some reports suggest that patients with SC-CDI may benefit from multiple serial FMT procedures (as opposed to a single FMT procedure, the standard for R-CDI) or from continuation of anti-CDI antibiotics concurrent with FMT (as opposed to prior cessation of antibiotics, the standard for R-CDI).[75-79] The decision to proceed with FMT in SC-CDI must take into account uncertainty regarding the optimal FMT strategy this population, as well as uncertainty regarding whether the expected benefit matches that of FMT for R-CDI.

In our experience, the alternative diagnosis of ischemic colitis should be considered in patients suspected of having SC-CDI, particularly if *C difficile* test results are equivocal. Ischemic colitis may manifest with symptoms similar to those of SC-CDI, and interaction between *C difficile* toxin and colonic ischemia may contribute to a more severe manifestation of CDI.[80-82] Colonoscopic examination and biopsy results may not help discriminate well between the two entities.[83,84] Although adverse events attributed to FMT have been rare, such events include perforated viscus, sepsis, and death in a patient with documented vascular disease.[85]

Early trials for FMT excluded patients with underlying bowel disease or impaired immune function, but case series suggest that FMT may be administered safely in patients with IBD; HIV; solid organ transplantation; leukemia or lymphoma; and recent exposure to antineoplastic or immunosuppressive agents, including monoclonal antibodies to B and T cells, antitumor necrosis factor, glucocorticoid, antimetabolite, and calcineurin-inhibitor medications.[86] Neutropenia remains a strict contraindication to FMT in most trials and observational studies and in our clinical practice. Patients with diseases or medications that may increase the theoretical risk of FMT-related infection should be counseled accordingly.

The FMT Procedure: Preparation, Administration, Expectations, and Alternatives

FMT requires healthy stool donors, infrastructure for the processing and storage of donated stool, and a mechanism for introducing stool from the healthy donor into the infected patient's small intestine or colon. Donor screening, stool processing, storage, and administration practices vary, but some standards have emerged, and ongoing trials will no doubt refine practice further.

FMT stool donors typically are screened for chronic medical conditions according to history and active infections according to serologic and stool testing. FMT screening guidelines published by leadership groups and centers with clinical experience generally concur that potential donors should provide a detailed history and be excluded if they report risk of transmitting an infectious agent (known HIV or viral hepatitis, illicit drug use, unprotected sex, sex with a high risk of HIV or hepatitis C virus acquisition, and so on), GI disease (IBD, IBS, GI malignancy, abdominal surgery, and so on), or other medical conditions or treatments that might impact the gut microbial community composition (obesity, metabolic syndrome, atopic disease, recent antibiotic use, and so on).

FDA guidance recommends its Full-Length Donor History Questionnaire (DHQ version 2.0) as a

framework for clinical screening.[87] The extent of recommended serologic and stool screening is more variable: Some authors favor abbreviated screening if the medical history is reassuring and if the donor is a family member or intimate of the recipient; others favor extensive screening, including standard blood-donor serologic screening and additional stool tests, for all donors.[22,47,88] A report of weight gain after FMT prompted recommendations for more rigorous biometric and serologic screening, including "normal BMI, testing of inflammatory markers, lipids, blood counts, electrolytes, and liver and renal function," as well as recommendations that healthy, volunteer donors be preferred over familial or intimate donors.[89,90] Table 1 summarizes a subset of the screening recommendations published by centers with clinical experience and by advisory groups.[22,47,50,88,91-95]

As standards for donor screening evolve, so do standards for FMT donation processing and storage. The first, to our knowledge, large randomized controlled trial of FMT used freshly donated stool.[27] Results from subsequent studies have supported the equivalent efficacy of frozen stool preparation.[41,42,44] Given the imperative for rigorous donor screening and the difficulty of timing donation, most institutions performing regular FMT have developed local banks of frozen stool preparations or work with outside stool banks such as OpenBiome to provide the same product.[48] Processing donated stool typically involves three steps: (1) homogenization and filtration to remove large particles, (2) concentration of bacteria via centrifugation, and (3) the addition of glycerol to permit frozen storage without bacterial cell lysis, which may impact the viability of bacterial cells and the efficacy of the FMT product.[47,96,97] The production of capsulized FMT for oral delivery also may involve the addition of a lipid emulsion so that capsules remain stable for ingestion.[98] A single FMT dose typically uses 50 g of fecal material, but a wide range of masses have been used, and one randomized controlled trial used 100 g of fecal material per dose.[24,27,41,42,44,47,99] It is expected that each gram of stool contains 10^{10} to 10^{12} bacterial cells.[100,101]

The FMT dose can be administered via the upper or lower GI tract. Upper GI delivery methods include oral capsules, nasogastric tubes, nasoduodenal tubes, and esophagogastroduodenoscopy; lower GI delivery methods include colonoscopy, sigmoidoscopy, and enema. Studies directly comparing delivery methods are scarce. Several authors have reported a 5% to 10% greater cure rate with lower GI delivery compared with the rate for upper GI delivery.[41,76,99,102-104] However, there are reports of cases refractory to lower GI delivery that responded to FMT via oral capsules,[105] and to date, to our knowledge, only a single, small trial has compared upper vs lower GI delivery directly.[41] Each approach has potential adverse effects: FMT delivered in the upper GI tract may not arrive promptly in the colon if CDI is accompanied by ileus, as is often true in SC-CDI. Delivery of FMT by means of lower GI endoscopy or enema may risk perforation if colonic inflammation is advanced, which also may be the case in SC-CDI.

Just as there is little consensus on the optimal mode of FMT delivery, there is little consensus on how best to prepare the recipient prior to FMT. Bowel lavage is known to reduce the luminal bacterial load significantly,[106] but whether it should be performed prior to FMT is uncertain. Large, randomized controlled trials that support the efficacy of FMT for R-CDI have been performed both with bowel lavage prior to FMT[23] and without.[44] It is typical to stop anti-CDI antibiotics 24 to 48 hours prior to the FMT procedure, but the impact of this timing on FMT success has not been evaluated rigorously, nor is there robust evidence to support which anti-CDI antibiotic should be used prior to FMT or at which dose. Some practitioners advocate the use of gastric acid suppression (typically a proton pump inhibitor) for 24 hours prior to upper GI FMT,[22] though this procedure has been used variably in randomized controlled trials.[27,41,42] Clinical evidence to determine whether bowel lavage and antacid treatment should be given is lacking, and the optimal pre-FMT bowel lavage, antibiotic, and antacid regimen is unknown.

In a broad array of observational studies and trials, as well as in our own experience, FMT cures $>$ 80% of R-CDI cases with a single dose. Repeat dosing typically achieves cure in $>$ 90% of patients with R-CDI.[3,27,31,40-44,74,107] In several reports and our own experience, the probability of successful FMT is reduced in inpatients, patients with SC-CDI, and patients with concurrent IBD, especially if diarrheal symptoms may be attributable to IBD itself and CDI was diagnosed by means of NAAT results alone.[108,109] Although case series results suggest that FMT has a role in the treatment of SC-CDI and CDI with concurrent IBD,[76,77,110] we presently lack adequate trial evidence to predict the probability of cure in patients with SC-CDI or CDI complicating IBD. A recent trial of FMT for

TABLE 1] Recommendations for FMT Donor Screening From Authors With Clinical Experience and From Advisory Panels

Donor Exclusion Criteria	Bakken et al/[22] 2011	Hamilton et al/[47] 2012	Health Canada/[92] 2015	Costello et al/[50] 2016	OpenBiome/[93] 2016
Clinical history					
Physician concern prompted by health history and lifestyle screen	+	+	+	+	+
Age < 18 or > 50 y	…	…	…	…	+
Age < 18 or > 65 y	…	…	…	+	+
Known HIV	+	+	+	+	+
Known HBV, hepatitis C virus	+	+	+	+	+
Exposure to HIV, viral hepatitis (within 12 mo)	+	+	+	+	+
High-risk sexual behavior	+	+	+	+	+
Use of illicit drugs	+	+	+	+	+
Tattoo or body piercing (within 6 mo)	+	+	+	+	+
Incarceration or history of incarceration	+	+	+	+	+
Known current infection (eg, URI)	+	+	+	+	+
Risk factors for vCJD	+	…	+	+	+
Travel to areas with endemic diarrheal diseases (within 6 mo)	+	+	+	+	+
History of IBD	+	+	+	+	+
History of IBS, idiopathic chronic constipation, or chronic diarrhea	+	+	+	+	+
History of GI malignancy or known polyposis	+	+	+	+	+
Antibiotics within 3 mo	+	+	+	+	+
Antibiotics within 6 mo	…	…	…	…	+
Major immunosuppressive medications	+	+	+	+	+
Systemic antineoplastic agents	+	+	+	+	+
Recent ingestion of a potential allergen (recipient with allergy)	+	…	+	+	+
History of major GI surgery (eg, gastric bypass)	+	+	…	+	+
Metabolic syndrome	+	+	…	+	+
Systemic autoimmunity (MS, SLE, and so on)	+	+	…	+	+
Atopic diseases (asthma and eczema, eosinophilic GI disorders)	+	+	…	+	+
Chronic pain syndromes (fibromyalgia, chronic fatigue)	+	…	…	+	+
Family history of colorectal carcinoma	…	…	…	+	+
Malnutrition (BMI < 18)	…	…	…	+	+
Risk for MDROs, including clinical work	…	…	…	…	+

(Continued)

TABLE 1] *(Continued)*

Donor Exclusion Criteria	Bakken et al/[22] 2011	Hamilton et al/[47] 2012	Health Canada/[92] 2015	Costello et al/[50] 2016	OpenBiome/[93] 2016
Stool testing					
Clostridium difficile toxin at PCR or EIA	+	+	+	+	+
Routine stool culture enteropathogens	+	+	+	+	+
Shiga toxin EIA	…	…	+	…	+
Fecal *Giardia* antigen	+	+	[a]	+	+
Fecal *Cryptosporidium* antigen	+	+	[a]	+	+
Acid-fast for *Cyclospora*, *Isospora*	+	…	[a]	+	+
Ova and parasites at microscopy	+	+	+	+	+
Helicobacter pylori fecal antigen	+	…	+	+	+
Rotavirus EIA or PCR	…	…	+	+	+
Norovirus PCR	…	…	+	+	+
Adenovirus EIA or PCR	…	…	+	+	+
Entamoeba histolytica PCR	…	…	+	+	…
Vancomycin-resistant *Enterococcus* screen	…	…	+	+	+
Extended-spectrum beta-lactamase screen	…	…	…	…	+
Carbapenemase-producing enterobacteriaceae screen	…	…	…	…	+
16S rRNA sequencing	…	…	…	…	+
Serologic and other testing					
HIV 1/2 antibody	+	+	+	+	+
HIV p24 antigen	…	…	+	+	+
HAV IgM	+	…	+	+	+
HBV surface antigen	+	+	+	+	+
HBV core antibody (IgG and IgM)	+	+	+	+	+
HBV surface antibody	+	+	+	+	+
HCV antibody	+	+	+	+	+
RPR	+	…	+	+	+
Treponemal antibody	+	…	+	…	+
HTLV 1/2 antibody	…	…	+	+	+
EBV IgG and IgM	…	…	…	+	…
CMV IgG and IgM	…	…	…	+	…
Strongyloides antibody	…	…	+	+	…
E histolytica antibody	…	…	+	+	…
H pylori antibody	…	…	+	+	…
Complete blood cell count	…	…	…	+	+
C-reactive protein	…	…	…	+	…
Erythrocyte sedimentation rate	…	…	…	+	…
Basic metabolic panel	…	…	…	+	…
Liver function tests	…	…	…	+	+
Fasting lipids	…	…	…	+	…
MRSA nasal culture	…	…	+	…	+
Malaria and babesiosis screen	…	…	+	…	…
Gonorrhea and chlamydia screen	…	…	+	…	…

(Continued)

TABLE 1] *(Continued)*

Donor Exclusion Criteria	Bakken et al/[22] 2011	Hamilton et al/[47] 2012	Health Canada/[92] 2015	Costello et al/[50] 2016	OpenBiome/[93] 2016
Other guidance					
Discretion to limit donor screening for intimates	Yes	No	Yes	NA	NA
Single donor or pooled stool product	Single	Single	Single	Single	Single

The plus sign (+) indicates specifically recommended screening or exclusion criteria. CMV = *Cytomegalovirus*; EBV = Epstein-Barr virus; EIA = enzyme immunoassay; FMT = fecal microbiota transplantation; HAV = hepatitis A virus; HBV = hepatitis B virus; HCV = hepatitis C virus; HTLV = human T-cell lymphotropic virus; IBD = inflammatory bowel disease; IBS = irritable bowel syndrome; MDRO = multidrug-resistant organism; MRSA = methicillin-resistant *Staphylococcus aureus*; MS = multiple sclerosis; NA = not applicable; PCR = polymerase chain reaction; rRNA = ribosomal RNA; RPR = rapid plasma reagin; SLE = systemic lupus erythematosus; URI = upper respiratory infection; vCJD = variant Creutzfeldt-Jakob disease
[a]Health Canada guidelines call for "protozoa" screening, but do not specify individual tests.

secondary prevention of CDI recurrence failed to demonstrate an advantage over prolonged use of oral vancomycin.[111]

It is essential to inform patients seeking FMT for R-CDI not only of the probability of treatment success but also of the symptoms to be expected after the procedure.[112-114] In the first 24 hours after FMT, bloating, flatulence, and loose stools are common. These symptoms typically resolve quickly, though some patients complain of bloating and flatulence for weeks. IBS following CDI and after FMT is well documented and may be characterized by lactose intolerance. Diarrhea may recur on isolated days, but patients whose FMTs are successful typically progress to formed stools over 1 to 2 weeks. Early FMT failure is far more common than late FMT failure: A recent cohort study found the rate of early (within 1 month) failure to be 18.6% and of late (1-3 months after FMT) failure to be 2.7%.[109] Failure typically manifests as recurrent diarrhea that progresses over several consecutive days. Such symptoms should prompt repeat stool testing for *C difficile*. Some practitioners advocate routine post-FMT stool testing, but we have avoided this practice given the uncertain significance of persistent positive NAAT results, as discussed.

The primary alternative to FMT for R-CDI is a prolonged antibiotic taper with pulsed dosing, which commonly is paired with a high-dose probiotic such as kefir.[71,115] The monoclonal antibody bezlotoxumab has been shown to reduce the risk of CDI recurrence when administered with standard antibiotic therapy for CDI, but its optimal use in secondary prevention of R-CDI has not yet been established.[116-118] The primary alternative to FMT for SC-CDI is surgery, either colectomy or diverting loop ileostomy with colonic lavage.[119] The details of these treatments are beyond the scope of this review.

Impact of FMT Beyond CDI

FMT is an effective means for treating CDI, particularly R-CDI. Its use for the treatment of other diseases is an area of active exploration. When advising FMT for the treatment of CDI in patients with other underlying bowel disease, it is essential to consider the potential for off-target FMT effects. In particular, FMT may impact IBD activity.

Whether FMT aids IBD control or provokes IBD exacerbation is an area of active debate, and at present no conclusive statement can be made on the balance between FMT's benefits and risks in this population. Several studies have been performed to evaluate the efficacy of FMT as a treatment for IBD, and they have produced conflicting results. Reports of FMT benefitting patients with IBD include remission rates as high as 27%,[120-122] but reports suggesting benefit have been matched by reports of adverse events that may be attributable to FMT, including IBD exacerbation and microscopic colitis.[108,123,124] The possibility of IBD exacerbation induced by FMT should highlight the importance of discriminating CDI from IBD.[68]

FMT from donors without recent antibiotic exposure also has been observed to reduce colonization by multidrug-resistant organisms (MDROs), and mechanisms by which FMT contributes to MDRO colonization resistance have been proposed.[125-131] This off-target effect appears purely beneficial, but its impact on subsequent risk for MDRO infection has not been studied thoroughly. Until the impact of FMT on the risk for subsequent MDRO infection is better characterized,

this potential benefit should not sway the decision on whether to proceed with FMT.

Infrastructure for Clinical FMT: Oversight, Billing, and Anticipated Changes

The greatest impediments to the broad dissemination of FMT for the treatment of R-CDI are uncertainty surrounding regulation by the FDA and the difficulty of obtaining reimbursement. Present regulation is not unduly onerous for FMT practitioners, but the FDA is reviewing its FMT guidance. The FDA considers FMT an investigational drug but declared in 2013 that it would exercise "enforcement discretion" and not require an Investigational New Drug (IND) application from physicians who perform FMT for the treatment of CDI that does not respond to anti-CDI antibiotics.[52] The consideration of FMT as a drug places it under the "Guidance for Industry: Enforcement Policy Regarding Investigational New Drugs"[132] rather than the regulations that the FDA uses for the oversight of blood transfusions (including donor screening, blood testing, donor deferral lists, quarantine, and regulations to address deficiencies), which might be more appropriate.

An IND application is required if FMT is to be performed for any indication other than CDI, and such treatment should be performed only in the setting of a clinical trial.[39,40] In March 2016, the FDA announced an intention to revise its FMT oversight, with specific consideration given to IND application requirements for large stool banks.[132] The revised policy has not yet been finalized.

Cost-effectiveness analyses have demonstrated substantial cost savings and increased efficacy of FMT for R-CDI, relative to those for oral vancomycin therapy.[133-135] Despite these data, in our experience, insurance reimbursement for FMT is not reliable, and reimbursement rates rarely cover the substantial cost of donor screening or purchase of the FMT dose from a stool bank. If FMT donation processing and dose administration are performed by the FMT practitioner, two Current Procedural Terminology codes can be billed: 44705 for the preparation of fecal microbiota for instillation (alternative for Medicare: G0455) and 44799 for the FMT instillation via naso- or oroenteric tube or enema. Esophagogastroduodenoscopy and colonoscopy should be billed as they would be normally. If the FMT material is obtained from a stool bank, it is recommended that code J3590 ("unclassified biologics") be used in lieu of 44705 and 44799, but no reimbursement is to be expected from this code.[136]

Conclusions

We have reviewed the clinical application of FMT for the treatment of CDI, paying particular attention to the care that must be taken in determining eligibility for FMT, the (evolving) present standards for FMT preparation and administration, the uncertainty of FMT regulation and difficulty of FMT billing, as well as emerging data on FMT effects beyond CDI. FMT has revolutionized the treatment of R-CDI, but much work remains to be done in defining the optimal FMT dose, as well as optimal protocols for donor screening, recipient preparation, and post-FMT monitoring. We remain in the early stages of establishing FMT's role in the treatment of SC-CDI, early CDI, and other bowel diseases. Given the benefits that FMT has demonstrated already, it is imperative that work to improve FMT efficacy and extend its benefits proceed rapidly.

Acknowledgments

Financial/nonfinancial disclosures: None declared.

Role of sponsors: The sponsor had no role in the design of the study, the collection and analysis of the data, or the preparation of the manuscript.

References

1. Bartlett JG, Gerding DN. Clinical recognition and diagnosis of *Clostridium difficile* infection. *Clin Infect Dis*. 2008;46(suppl 1): S12-S18.
2. Cohen SH, Gerding DN, Johnson S, et al; and the Society for Healthcare Epidemiology of America; Infectious Diseases Society of America. Clinical practice guidelines for *Clostridium difficile* infection in adults: 2010 update by the society for healthcare epidemiology of America (SHEA) and the infectious diseases society of America (IDSA). *Infect Control Hosp Epidemiol*. 2010;31(5):431-455.
3. Surawicz CM, Brandt LJ, Binion DG, et al. Guidelines for diagnosis, treatment, and prevention of *Clostridium difficile* infections. *Am J Gastroenterol*. 2013;108(4):478-498.
4. Dubberke ER, Carling P, Carrico R, et al. Strategies to prevent *Clostridium difficile* infections in acute care hospitals: 2014 update. *Infect Control Hosp Epidemiol*. 2014;35(6):628-645.
5. Eyre DW, Cule ML, Wilson DJ, et al. Diverse sources of *C. difficile* infection identified on whole-genome sequencing. *N Engl J Med*. 2013;369(13):1195-1205.
6. Kumar N, Miyajima F, He M, et al. Genome-based infection tracking reveals dynamics of *Clostridium difficile* transmission and disease recurrence. *Clin Infect Dis*. 2016;62(6):746-752.
7. Seekatz AM, Young VB. *Clostridium difficile* and the microbiota. *J Clin Invest*. 2014;124(10):4182-4189.
8. Schubert AM, Rogers MAM, Ring C, et al. Microbiome data distinguish patients with *Clostridium difficile* infection and non-*C. difficile*-associated diarrhea from healthy controls. *MBio*. 2014;5(3): e01021-14.
9. Koenigsknecht MJ, Theriot CM, Bergin IL, Schumacher CA, Schloss PD, Young VB. Dynamics and establishment of *Clostridium difficile* infection in the murine gastrointestinal tract. *Infect Immun*. 2015;83(3):934-941.

10. Schubert AM, Sinani H, Schloss PD. Antibiotic-induced alterations of the murine gut microbiota and subsequent effects on colonization resistance against. *Clostridium difficile. MBio*. 2015;6(4):e00974-15.

11. Buffie CG, Bucci V, Stein RR, et al. Precision microbiome reconstitution restores bile acid mediated resistance to. *Clostridium difficile. Nature*. 2015;517(7533):205-208.

12. Lewis BB, Carter RA, Pamer EG. Bile acid sensitivity and in vivo virulence of clinical *Clostridium difficile* isolates. *Anaerobe*. 2016;41:32-36.

13. Manges AR, Labbe A, Loo VG, et al. Comparative metagenomic study of alterations to the intestinal microbiota and risk of nosocomial *Clostridium difficile*-associated disease. *J Infect Dis*. 2010;202(12):1877-1884.

14. Vincent C, Stephens DA, Loo VG, et al. Reductions in intestinal Clostridiales precede the development of nosocomial *Clostridium difficile* infection. *Microbiome*. 2013;1(1):18.

15. Shreiner AB, Kao JY, Young VB. The gut microbiome in health and in disease. *Curr Opin Gastroenterol*. 2015;31(1):69-75.

16. Lee YJ, Arguello EP, Jenq RR, et al. Protective factors in the intestinal microbiome against *Clostridium difficile* infection in recipients of allogeneic hematopoietic stem cell transplantation. *J Infect Dis*. 2017;215(7):1117-1123.

17. Loo VG, Bourgault AM, Poirier L, et al. Host and pathogen factors for *Clostridium difficile* infection and colonization. *N Engl J Med*. 2011;365(18):1693-1703.

18. Kong LY, Dendukuri N, Schiller I, et al. Predictors of asymptomatic *Clostridium difficile* colonization on hospital admission. *Am J Infect Control*. 2015;43(3):248-253.

19. Leffler DA, Lamont JT. *Clostridium difficile* infection. *N Engl J Med*. 2015;372(16):1539-1548.

20. Arriola V, Tischendorf J, Musuuza J, Barker A, Rozelle JW, Safdar N. Assessing the risk of hospital-acquired *Clostridium difficile* infection with proton pump inhibitor use: a meta-analysis. *Infect Control Hosp Epidemiol*. 2016;37(12):1408-1417.

21. Aas J, Gessert CE, Bakken JS. Recurrent *Clostridium difficile* colitis: case series involving 18 patients treated with donor stool administered via a nasogastric tube. *Clin Infect Dis*. 2003;36(5):580-585.

22. Bakken JS, Borody T, Brandt LJ, et al. Treating *Clostridium difficile* infection with fecal microbiota transplantation. *Clin Gastroenterol Hepatol*. 2011;9(12):1044-1049.

23. Brandt LJ. FMT: first step in a long journey. *Am J Gastroenterol*. 2013;108(8):1367-1368.

24. Cammarota G, Ianiro G, Gasbarrini A; and the Fecal Microbiota Transplantation Workgroup. Fecal microbiota transplantation for the treatment of *Clostridium difficile* infection: a systematic review. *J Clin Gastroenterol*. 2014;48(8):693-702.

25. Brandt LJ. Fecal microbiota transplant: respice, adspice, prospice. *J Clin Gastroenterol*. 2015;49(suppl 1):S65-S68.

26. Kelly CR, Kahn S, Kashyap P, et al. Update on fecal microbiota transplantation 2015: indications, methodologies, mechanisms, and outlook. *Gastroenterology*. 2015;149(1):223-237.

27. van Nood E, Vrieze A, Nieuwdorp M, et al. Duodenal infusion of donor feces for recurrent. *Clostridium difficile. N Engl J Med*. 2013;368(5):407-415.

28. Song Y, Garg S, Girotra M, et al. Microbiota dynamics in patients treated with fecal microbiota transplantation for recurrent *Clostridium difficile* infection. *PLoS One*. 2013;8(11):e81330.

29. Hamilton MJ, Weingarden AR, Unno T, Khoruts A, Sadowsky MJ. High-throughput DNA sequence analysis reveals stable engraftment of gut microbiota following transplantation of previously frozen fecal bacteria. *Gut Microbes*. 2013;4(2):125-135.

30. Seekatz AM, Aas J, Gessert CE, et al. Recovery of the gut microbiome following fecal microbiota transplantation. *MBio*. 2014;5(3):e00893-14.

31. Dutta SK, Girotra M, Garg S, et al. Efficacy of combined jejunal and colonic fecal microbiota transplantation for recurrent *Clostridium difficile* infection. *Clin Gastroenterol Hepatol*. 2014;12(9):1572-1576.

32. Weingarden A, González A, Vázquez-Baeza Y, et al. Dynamic changes in short- and long-term bacterial composition following fecal microbiota transplantation for recurrent *Clostridium difficile* infection. *Microbiome*. 2015;3:10.

33. Khanna S, Montassier E, Schmidt B, et al. Gut microbiome predictors of treatment response and recurrence in primary *Clostridium difficile* infection. *Aliment Pharmacol Ther*. 2016;44(7):715-727.

34. Jalanka J, Mattila E, Jouhten H, et al. Long-term effects on luminal and mucosal microbiota and commonly acquired taxa in faecal microbiota transplantation for recurrent *Clostridium difficile* infection. *BMC Med*. 2016;14(1):155.

35. Staley C, Kelly CR, Brandt LJ, Khoruts A, Sadowsky MJ. Complete microbiota engraftment is not essential for recovery from recurrent *Clostridium difficile* infection following fecal microbiota transplantation. *MBio*. 2016;7(6):e01965-16.

36. Li SS, Zhu A, Benes V, et al. Durable coexistence of donor and recipient strains after fecal microbiota transplantation. *Science*. 2016;352(6285):586-589.

37. Bojanova DP, Bordenstein SR. Fecal transplants: what is being transferred? *PLoS Biol*. 2016;14(7):e1002503.

38. Lessa FC, Mu Y, Bamberg WM, et al. Burden of *Clostridium difficile* infection in the United States. *N Engl J Med*. 2015;372(9):825-834.

39. Gerding DN, Lessa FC. The epidemiology of *Clostridium difficile* infection inside and outside health care institutions. *Infect Dis Clin North Am*. 2015;29(1):37-50.

40. Petrof EO, Gloor GB, Vanner SJ, et al. Stool substitute transplant therapy for the eradication of *Clostridium difficile* infection: "RePOOPulating" the gut. *Microbiome*. 2013;1(1):3.

41. Youngster I, Sauk J, Pindar C, et al. Fecal microbiota transplant for relapsing *Clostridium difficile* infection using a frozen inoculum from unrelated donors: a randomized, open-label, controlled pilot study. *Clin Infect Dis*. 2014;58(11):1515-1522.

42. Youngster I, Russell GH, Pindar C, Ziv-Baran T, Sauk J, Hohmann EL. Oral, capsulized, frozen fecal microbiota transplantation for relapsing *Clostridium difficile* infection. *JAMA*. 2014;312(17):1772-1778.

43. Cammarota G, Masucci L, Ianiro G, et al. Randomised clinical trial: faecal microbiota transplantation by colonoscopy vs. vancomycin for the treatment of recurrent *Clostridium difficile* infection. *Aliment Pharmacol Ther*. 2015;41(9):835-843.

44. Lee CH, Steiner T, Petrof EO, et al. Frozen vs fresh fecal microbiota transplantation and clinical resolution of diarrhea in patients with recurrent *Clostridium difficile* infection: a randomized clinical trial. *JAMA*. 2016;315(2):142-149.

45. Kelly CR, Khoruts A, Staley C, et al. Effect of fecal microbiota transplantation on recurrence in multiply recurrent *Clostridium difficile* infection: a randomized trial. *Ann Intern Med*. 2016;165(9):609-616.

46. Rao K, Young VB. Fecal microbiota transplantation for the management of *Clostridium difficile* infection. *Infect Dis Clin North Am*. 2015;29(1):109-122.

47. Hamilton MJ, Weingarden AR, Sadowsky MJ, Khoruts A. Standardized frozen preparation for transplantation of fecal microbiota for recurrent *Clostridium difficile* infection. *Am J Gastroenterol*. 2012;107(5):761-767.

48. Kazerouni A, Burgess J, Burns LJ, Wein LM. Optimal screening and donor management in a public stool bank. *Microbiome*. 2015;3(1):75.

49. Khoruts A, Sadowsky MJ, Hamilton MJ. Development of fecal microbiota transplantation suitable for mainstream medicine. *Clin Gastroenterol Hepatol*. 2015;13(2):246-250.

50. Costello SP, Tucker EC, La Brooy J, Schoeman MN, Andrews JM. Establishing a fecal microbiota transplant service for the treatment of *Clostridium difficile* infection. *Clin Infect Dis*. 2016;62(7):908-914.

51. Moore T, Rodriguez A, Bakken JS. Fecal microbiota transplantation: a practical update for the infectious disease specialist. *Clin Infect Dis*. 2014;58(4):541-545.

52. Kelly CR, Kunde SS, Khoruts A. Guidance on preparing an investigational new drug application for fecal microbiota transplantation studies. *Clin Gastroenterol Hepatol*. 2014;12(2):283-288.

53. Polage CR, Gyorke CE, Kennedy MA, et al. Overdiagnosis of *Clostridium difficile* infection in the molecular test era. *JAMA Intern Med*. 2015;175(11):1792-1801.

54. Polage CR, Turkiewicz JV, Cohen SH. The never-ending struggle with laboratory testing for *Clostridium difficile* infection. *J Comp Eff Res*. 2016;5(2):113-116.

55. Gould CV, Edwards JR, Cohen J, et al; and the Clostridium difficile Infection Surveillance Investigators, Centers for Disease Control and Prevention. Effect of nucleic acid amplification testing on population-based incidence rates of *Clostridium difficile* infection. *Clin Infect Dis*. 2013;57(9):1304-1307.

56. Crobach MJT, Planche T, Eckert C, et al. European Society of Clinical Microbiology and Infectious Diseases: update of the diagnostic guidance document for *Clostridium difficile* infection. *Clin Microbiol Infect*. 2016;22(suppl 4):S63-S81.

57. Owens RC Jr, Donskey CJ, Gaynes RP, Loo VG, Muto CA. Antimicrobial-associated risk factors for *Clostridium difficile* infection. *Clin Infect Dis*. 2008;46(suppl 1):S19-S31.

58. Khanna S, Pardi DS, Aronson SL, et al. The epidemiology of community-acquired *Clostridium difficile* infection: a population-based study. *Am J Gastroenterol*. 2012;107(1):89-95.

59. Freedberg DE, Salmasian H, Friedman C, Abrams JA. Proton pump inhibitors and risk for recurrent *Clostridium difficile* infection among inpatients. *Am J Gastroenterol*. 2013;108(11):1794-1801.

60. Chitnis AS, Holzbauer SM, Belflower RM, et al. Epidemiology of community-associated *Clostridium difficile* infection, 2009 through 2011. *JAMA Intern Med*. 2013;173(14):1359-1367.

61. Lessa FC, Mu Y, Winston LG, et al. Determinants of *Clostridium difficile* infection incidence across diverse United States geographic locations. *Open Forum Infect Dis*. 2014;1(2):ofu048.

62. Jackson MA, Goodrich JK, Maxan ME, et al. Proton pump inhibitors alter the composition of the gut microbiota. *Gut*. 2016;65(5):749-756.

63. Khanna S, Pardi DS. Clinical implications of antibiotic impact on gastrointestinal microbiota and *Clostridium difficile* infection. *Expert Rev Gastroenterol Hepatol*. 2016;10(10):1145-1152.

64. Tariq R, Singh S, Gupta A, Pardi DS, Khanna S. Association of gastric acid suppression with recurrent *Clostridium difficile* infection: a systematic review and meta-analysis. *JAMA Intern Med*. 2017;177(6):784-791.

65. Kwon JH, Reske KA, Hink T, Burnham CA, Dubberke ER. Evaluation of correlation between pretest probability for *Clostridium difficile* infection and *Clostridium difficile* enzyme immunoassay results. *J Clin Microbiol*. 2017;55(2):596-605.

66. Bartlett JG. Antibiotic-associated diarrhea. *N Engl J Med*. 2002;346(5):334-339.

67. Hohmann EL, Ananthakrishnan AN, Deshpande V. Case Records of the Massachusetts General Hospital: case 25-2014—a 37-year-old man with ulcerative colitis and bloody diarrhea. *N Engl J Med*. 2014;371(7):668-675.

68. Jackson M, Olefson S, Machan JT, Kelly CR. A high rate of alternative diagnoses in patients referred for presumed *Clostridium difficile* infection. *J Clin Gastroenterol*. 2016;50(9):742-746.

69. Larson AM, Fung AM, Fang FC. Evaluation of tcdB real-time PCR in a three-step diagnostic algorithm for detection of toxigenic *Clostridium difficile*. *J Clin Microbiol*. 2010;48(1):124-130.

70. Tenover FC, Novak-Weekley S, Woods CW, et al. Impact of strain type on detection of toxigenic *Clostridium difficile*: comparison of molecular diagnostic and enzyme immunoassay approaches. *J Clin Microbiol*. 2010;48(10):3719-3724.

71. McFarland LV, Elmer GW, Surawicz CM. Breaking the cycle: treatment strategies for 163 cases of recurrent *Clostridium difficile* disease. *Am J Gastroenterol*. 2002;97(7):1769-1775.

72. Tang-Feldman Y, Mayo S, Silva J Jr, Cohen SH. Molecular analysis of *Clostridium difficile* strains isolated from 18 cases of recurrent *Clostridium difficile*-associated diarrhea. *J Clin Microbiol*. 2003;41(7):3413-3414.

73. Seekatz AM, Rao K, Santhosh K, Young VB. Dynamics of the fecal microbiome in patients with recurrent and nonrecurrent *Clostridium difficile* infection. *Genome Med*. 2016;8(1):47.

74. Debast SB, Bauer MP, Kuijper EJ; and the European Society of Clinical Microbiology and Infectious Diseases. European Society of Clinical Microbiology and Infectious Diseases: update of the treatment guidance document for *Clostridium difficile* infection. *Clin Microbiol Infect*. 2014;20(suppl 2):1-26.

75. Weingarden AR, Hamilton MJ, Sadowsky MJ, Khoruts A. Resolution of severe *Clostridium difficile* infection following sequential fecal microbiota transplantation. *J Clin Gastroenterol*. 2013;47(8):735-737.

76. Agrawal M, Aroniadis OC, Brandt LJ, et al. The long-term efficacy and safety of fecal microbiota transplant for recurrent, severe, and complicated *Clostridium difficile* infection in 146 elderly individuals. *J Clin Gastroenterol*. 2016;50(5):403-407.

77. Aroniadis OC, Brandt LJ, Greenberg A, et al. Long-term follow-up study of fecal microbiota transplantation for severe and/or complicated *Clostridium difficile* infection: a multicenter experience. *J Clin Gastroenterol*. 2016;50(5):398-402.

78. Fischer M, Sipe BW, Rogers NA, et al. Faecal microbiota transplantation plus selected use of vancomycin for severe-complicated *Clostridium difficile* infection: description of a protocol with high success rate. *Aliment Pharmacol Ther*. 2015;42(4):470-476.

79. Pecere S, Sabatelli M, Fantoni M, Ianiro G, Gasbarrini A, Cammarota G. Letter: faecal microbiota transplantation in combination with fidaxomicin to treat severe complicated recurrent *Clostridium difficile* infection [letter]. *Aliment Pharmacol Ther*. 2015;42(8):1030.

80. Dial S. *Clostridium difficile* colitis: a marker for ischemic colitis? [letter]. *CMAJ*. 2004;171(11):1326.

81. Dallal RM, Harbrecht BG, Boujoukas AJ, et al. Fulminant *Clostridium difficile*: an underappreciated and increasing cause of death and complications. *Ann Surg*. 2002;235(3):363-372.

82. Mulki R, Baumann AJ, Alnabelsi T, et al. Body mass index greater than 35 is associated with severe *Clostridium difficile* infection. *Aliment Pharmacol Ther*. 2017;45(1):75-81.

83. Dignan CR, Greenson JK. Can ischemic colitis be differentiated from *C difficile* colitis in biopsy specimens? *Am J Surg Pathol*. 1997;21(6):706-710.

84. Tang DM, Urrunaga NH, De Groot H, von Rosenvinge EC, Xie G, Ghazi LJ. Pseudomembranous colitis: not always caused by *Clostridium difficile*. *Case Rep Med*. 2014;2014:812704.

85. Solari PR, Fairchild PG, Noa LJ, Wallace MR. Tempered enthusiasm for fecal transplant [letter]. *Clin Infect Dis*. 2014;59(2):319.

86. Kelly CR, Ihunnah C, Fischer M, et al. Fecal microbiota transplant for treatment of *Clostridium difficile* infection in immunocompromised patients. *Am J Gastroenterol*. 2014;109(7):1065-1071.

87. US Department of Health and Human Services Food and Drug Administration Center for Biologics Evaluation and Research. Implementation of acceptable full-length and abbreviated donor history questionnaires and accompanying materials for use in screening donors of blood and blood components: guidance for industry. https://www.fda.gov/downloads/regulatoryinformation/guidances/ucm273685.pdf. Accessed September 22, 2017.

88. Rohlke F, Stollman N. Fecal microbiota transplantation in relapsing *Clostridium difficile* infection. *Therap Adv Gastroenterol*. 2012;5(6):403-420.

89. Alang N, Kelly CR. Weight gain after fecal microbiota transplantation. *Open Forum Infect Dis*. 2015;2(1):ofv004.

90. Weil AA, Hohmann EL. Fecal microbiota transplant: benefits and risks. *Open Forum Infect Dis*. 2015;2(1):ofv005.

91. Mattila E, Uusitalo-Seppälä R, Wuorela M, et al. Fecal transplantation, through colonoscopy, is effective therapy for recurrent *Clostridium difficile* infection. *Gastroenterology*. 2012;142(3):490-496.

92. Government of Canada. Guidance document: fecal microbiota therapy used in the treatment of *Clostridium difficile* infection not responsive to standard therapies. https://www.canada.ca/en/health-canada/services/drugs-health-products/public-involvement-consultations/biologics-radiopharmaceuticals-genetic-therapies/guidance-document-regulation-fecal-microbiota-therapy.html. Accessed September 22, 2017.

93. OpenBiome. Quality & Safety Program. https://static1.squarespace.com/static/50e0c29ae4b0a05702af7e6a/t/59304bdf3e00be24622791fb/1496337376745/The+OpenBiome+Quality+%26+Safety+Program.pdf. Accessed September 1, 2017.

94. Tariq R, Weatherly R, Kammer P, Pardi DS, Khanna S. Donor screening experience for fecal microbiota transplantation in patients with recurrent *C. difficile* infection [published online ahead of print December 14, 2016]. *J Clin Gastroenterol*. https://doi.org/10.1097/MCG.0000000000000768.

95. Sokol H, Galperine T, Kapel N, et al; and the French Group of Faecal microbiota Transplantation. Faecal microbiota transplantation in recurrent *Clostridium difficile* infection: recommendations from the French Group of Faecal microbiota Transplantation. *Dig Liver Dis*. 2016;48(3):242-247.

96. Craven LJ, Gloor G, Reid G, Silverman M, Burton JP. What are the optimal long-term storage conditions for donor samples used in fecal microbiota transplantation? Abstract presented at: ASM Microbe 2017 meeting; June 3, 2017; New Orleans, LA.

97. Paterson AS, Kissoon M, Surangiwala S, Willey BM, Hota S, Poutanen SM. Determination of optimal duration and conditions for long-term storage of fecal filtrate samples. Abstract presented at: ASM Microbe 2017 meeting; June 4, 2017; New Orleans, LA.

98. OpenBiome. FMT capsule G3 clinical primer. https://www.openbiome.org/s/G3-Capsules-Clinical-Primer. Accessed September 22, 2017.

99. Kassam Z, Lee CH, Yuan Y, Hunt RH. Fecal microbiota transplantation for *Clostridium difficile* infection: systematic review and meta-analysis. *Am J Gastroenterol*. 2013;108(4):500-508.

100. Simon GL, Gorbach SL. Intestinal flora in health and disease. *Gastroenterology*. 1984;86(1):174-193.

101. Franks AH, Harmsen HJM, Raangs GC, Jansen GJ, Schut F, Welling GW. Variations of bacterial populations in human feces measured by fluorescent in situ hybridization with group-specific 16S rRNA-targeted oligonucleotide probes. *Appl Environ Microbiol*. 1998;64(9):3336-3345.

102. Gough E, Shaikh H, Manges AR. Systematic review of intestinal microbiota transplantation (fecal bacteriotherapy) for recurrent *Clostridium difficile* infection. *Clin Infect Dis*. 2011;53(10):994-1002.

103. Postigo R, Kim JH. Colonoscopic versus nasogastric fecal transplantation for the treatment of *Clostridium difficile* infection: a review and pooled analysis. *Infection*. 2012;40(6):643-648.

104. Sofi AA, Silverman AL, Khuder S, Garborg K, Westerink JMA, Nawras A. Relationship of symptom duration and fecal bacteriotherapy in *Clostridium difficile* infection: pooled data analysis and a systematic review. *Scand J Gastroenterol*. 2013;48(3):266-273.

105. Stollman N, Smith M, Giovanelli A, et al. Frozen encapsulated stool in recurrent *Clostridium difficile*: exploring the role of pills in the treatment hierarchy of fecal microbiota transplant nonresponders. *Am J Gastroenterol*. 2015;110(4):600-601.

106. Jalanka J, Salonen A, Salojärvi J, et al. Effects of bowel cleansing on the intestinal microbiota. *Gut*. 2015;64(10):1562-1568.

107. Youngster I, Mahabamunuge J, Systrom HK, et al. Oral, frozen fecal microbiota transplant (FMT) capsules for recurrent *Clostridium difficile* infection. *BMC Med*. 2016;14(1):134.

108. Khoruts A, Rank KM, Newman KM, et al. Inflammatory bowel disease affects the outcome of fecal microbiota transplantation for recurrent *Clostridium difficile* infection. *Clin Gastroenterol Hepatol*. 2016;14(10):1433-1438.

109. Fischer M, Kao D, Mehta SR, et al. Predictors of early failure after fecal microbiota transplantation for the therapy of *Clostridium difficile* infection: a multicenter study. *Am J Gastroenterol*. 2016;111(7):1024-1031.

110. Fischer M, Kao D, Kelly C, et al. Fecal microbiota transplantation is safe and efficacious for recurrent or refractory *Clostridium difficile* infection in patients with inflammatory bowel disease. *Inflamm Bowel Dis*. 2016;22(10):2402-2409.

111. Hota SS, Sales V, Tomlinson G, et al. Oral vancomycin followed by fecal transplantation versus tapering oral vancomycin treatment for recurrent *Clostridium difficile* infection: an open-label, randomized controlled trial. *Clin Infect Dis*. 2017;64(3):265-271.

112. Bokek-Cohen Y, Ravitsky V. Cultural and personal considerations in informed consent for fecal microbiota transplantation [abstract]. *Am J Bioeth*. 2017;17(5):55-57.

113. Ma Y, Liu J, Rhodes C, Nie Y, Zhang F. Ethical issues in fecal microbiota transplantation in practice. *Am J Bioeth*. 2017;17(5):34-45.

114. Bunnik EM, Aarts N, Chen LA. Physicians must discuss potential long-term risks of fecal microbiota transplantation to ensure informed consent. *Am J Bioeth*. 2017;17(5):61-63.

115. Bakken JS. Staggered and tapered antibiotic withdrawal with administration of kefir for recurrent *Clostridium difficile* infection. *Clin Infect Dis*. 2014;59(6):858-861.

116. Lowy I, Molrine DC, Leav BA, et al. Treatment with monoclonal antibodies against *Clostridium difficile* toxins. *N Engl J Med*. 2010;362(3):197-205.

117. Gupta SB, Mehta V, Dubberke ER, et al. Antibodies to toxin B are protective against *Clostridium difficile* infection recurrence. *Clin Infect Dis*. 2016;63(6):730-734.

118. Cornely OA, Vehreschild MJGT. Where to place the new treatments for *Clostridium difficile* infection? [editorial commentary]. *Clin Infect Dis*. 2016;63(6):735-736.

119. Neal MD, Alverdy JC, Hall DE, Simmons RL, Zuckerbraun BS. Diverting loop ileostomy and colonic lavage: an alternative to total abdominal colectomy for the treatment of severe, complicated *Clostridium difficile* associated disease. *Ann Surg*. 2011;254(3):423-427.

120. Moayyedi P, Surette MG, Kim PT, et al. Fecal microbiota transplantation induces remission in patients with active ulcerative colitis in a randomized controlled trial. *Gastroenterology*. 2015;149(1):102.e6-109.e6.

121. Jacob V, Crawford C, Cohen-Mekelburg S, et al. Single delivery of high-diversity fecal microbiota preparation by colonoscopy is safe and effective in increasing microbial diversity in active ulcerative colitis. *Inflamm Bowel Dis*. 2017;23(6):903-911.

122. Paramsothy S, Kamm MA, Kaakoush NO, et al. Multidonor intensive faecal microbiota transplantation for active ulcerative colitis: a randomised placebo-controlled trial. *Lancet*. 2017;389(10075):1218-1228.

123. Grinspan AM, Kelly CR. Fecal microbiota transplantation for ulcerative colitis: not just yet. *Gastroenterology*. 2015;149(1):15-18.

124. Tariq R, Smyrk T, Pardi DS, Tremaine WJ, Khanna S. New-onset microscopic colitis in an ulcerative colitis patient after fecal microbiota transplantation. *Am J Gastroenterol*. 2016;111(5):751-752.

125. Singh R, van Nood E, Nieuwdorp M, et al. Donor feces infusion for eradication of Extended Spectrum beta-Lactamase producing *Escherichia coli* in a patient with end stage renal disease. *Clin Microbiol Infect*. 2014;20(11):O977-O978.

126. Crum-Cianflone NF, Sullivan E, Ballon-Landa G. Fecal microbiota transplantation and successful resolution of multidrug-resistant-organism colonization. *J Clin Microbiol*. 2015;53(6):1986-1989.

127. Borody T, Fischer M, Mitchell S, Campbell J. Fecal microbiota transplantation in gastrointestinal disease: 2015 update and the

road ahead. *Expert Rev Gastroenterol Hepatol*. 2015;9(11):1379-1391.

128. Millan B, Park H, Hotte N, et al. Fecal microbial transplants reduce antibiotic-resistant genes in patients with recurrent *Clostridium difficile* infection. *Clin Infect Dis*. 2016;62(12):1479-1486.

129. Biliński J, Grzesiowski P, Muszyński J, et al. Fecal microbiota transplantation inhibits multidrug-resistant gut pathogens: preliminary report performed in an immunocompromised host. *Arch Immunol Ther Exp (Warsz)*. 2016;64(3):255-258.

130. Araos R, Tai AK, Snyder GM, Blaser MJ, D'Agata EMC. Predominance of *Lactobacillus* spp. among patients who do not acquire multidrug-resistant organisms. *Clin Infect Dis*. 2016;63(7):937-943.

131. Bilinski J, Grzesiowski P, Sorensen N, et al. Fecal microbiota transplantation in patients with blood disorders inhibits gut colonization with antibiotic-resistant bacteria: results of a prospective, single-center study. *Clin Infect Dis*. 2017;65(3):364-370.

132. Food and Drug Administration Center for Biologics Evaluation and Research. Enforcement policy regarding investigational new drug requirements for use of fecal microbiota for transplantation to treat *Clostridium difficile* infection not responsive to standard therapies: draft guidance for industry, March 2016. https://www.fda.gov/downloads/biologicsbloodvaccines/guidancecomplianceregulatoryinformation/guidances/vaccines/ucm488223.pdf. Accessed September 1, 2017

133. Konijeti GG, Sauk J, Shrime MG, Gupta M, Ananthakrishnan AN. Cost-effectiveness of competing strategies for management of recurrent *Clostridium difficile* infection: a decision analysis. *Clin Infect Dis*. 2014;58(11):1507-1514.

134. Varier RU, Biltaji E, Smith KJ, et al. Cost-effectiveness analysis of fecal microbiota transplantation for recurrent *Clostridium difficile* infection. *Infect Control Hosp Epidemiol*. 2015;36(4):438-444.

135. Baro E, Galperine T, Denies F, et al. Cost-effectiveness analysis of five competing strategies for the management of multiple recurrent community-onset *Clostridium difficile* infection in France. *PLoS One*. 2017;12(1):e0170258.

136. OpenBiome. The OpenBiome billing guide: a guide to physician and hospital billing for using OpenBiome fecal microbiota preparations. https://static1.squarespace.com/static/50e0c29ae4b0a05702af7e6a/t/578e3711579fb3d7cbe44eaa/1468938001601/Billing+Guide.pdf. Accessed September 22, 2017.