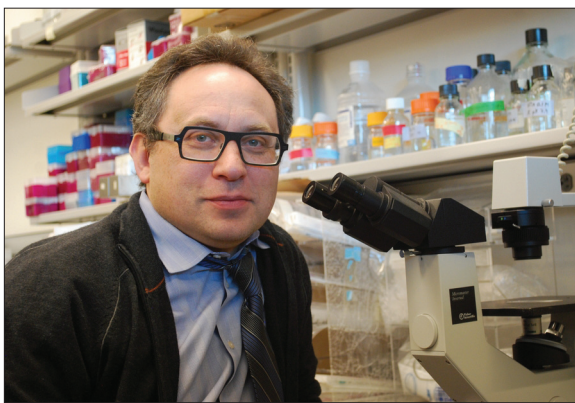


## INTERVIEW

# Fecal Microbiota Transplantation: An Interview With Alexander Khoruts

Steve LeBeau; Alexander Khoruts, MD



Dr Alexander Khoruts in his lab at the University of Minnesota.

Photo credit: Steve LeBeau.

**A** new, highly active sector of therapeutics in the form of fecal microbiota transplantation (FMT) has emerged in the last several years based on novel paradigms in medicine that are challenging to the regulators such as the US Food and Drug Administration (FDA). Dr Alexander Khoruts, associate professor of medicine at the University of Minnesota, Minneapolis, one of the leading researchers on FMT, discusses his work, the current regulatory challenges, and related topics.

**Mr LeBeau:** How did you become involved in studying FMT?

**Dr Khoruts:** I am a physician-scientist. My clinical specialty is gastroenterology, but I also had been trained as a basic immunologist. Until 2010, my major research focus has been on T cell biology in mouse models. However, for a number of years I have been following literature on the roles commensal microbes play in our bodies and in particular a series of papers from Dr Jeffrey Gordon's at Washington University School of Medicine, linking energy metabolism to activity of distal gut microbial communities.<sup>1-4</sup> I realized that the new DNA sequencing and computational technologies that enabled this new research of microbe-host interactions was revolutionary in Medicine. I believed we were witnessing discovery of a new organ in the human body, and this organ was central to my own clinical subspecialty and envied young doctors in training who could choose to start their research careers in this field. I did start thinking of ways to transition my focus at midcareer.

The opportunity presented itself in the form of a patient in 2008. The patient had refractory *Clostridium*

*difficile* infection (CDI) that could not be eradicated with antibiotics. The infection was triggered by antibiotics used to treat her for pneumonia and given prophylactically for back surgery. It has long been suspected that normal bacteria in the colon are able to protect the human host against CDI, and medical use of antibiotics makes patients vulnerable to this infection by killing normal bacteria in the gut. The problem can become very challenging in some patients because antibiotics used to treat CDI do not eliminate *C difficile* spores. In these patients, symptoms promptly return once anti-CDI antibiotics are stopped. The treatment perpetuates the problem. Ultimately it turns into the recurrent CDI syndrome (RCDI), a condition of indefinite infection cycles that cannot be eradicated with antibiotics alone.<sup>5</sup>

The patient was a 64-year-old woman who had recurrent CDI for approximately eight months before she was referred to see me in clinic. At that point, she had diarrheal stools every 15 minutes, round the clock, lived in diapers, and had lost approximately 60 pounds. She had taken multiple courses of antibiotics without success to eradicate the infection. In fact, her case was especially unusual in that she did not respond even temporarily to vancomycin, the most commonly used antibiotic approved for CDI. Her primary care physician, frustrated with therapeutic failures, referred her to see me, specifically for a stool transplant. At the time, this procedure was an oddity, sporadically described in isolated case reports. It certainly was not an established procedure at the University or anywhere in the Twin Cities, although a team in Duluth, Minnesota, has been doing them for several decades.

In Western medicine, fecal enema was originally reported by Dr Ben Eiseman and his team at the University of Colorado in 1958 as a cure for pseudomembranous colitis,<sup>6</sup> which we now recognize to be a severe form of CDI. At that time pseudomembranous colitis was not yet linked to *C difficile*. However, the researchers recognized that the problem was related to antibiotic use, probably due to suppression of normal bacteria in the colon. The clinical condition they described had 75% mortality at the time, but their patients literally walked out of the hospital after just a couple of days. Soon after that vancomycin was discovered to be effective in treatment of this infection, and stool transplants became very rare. Between 1958 and 1989 or so there were maybe 20 case reports. However, in the early 1990s we started seeing more aggressive forms of CDI. More virulent strains of *C difficile* emerged that produce greater quantities of toxins, had broader antibiotic resistance, and sporulated more efficiently.

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By the 2000s, CDI has become epidemic and more deadly. The US Centers for Disease Control and Prevention estimates this infection kills about 30,000 people per year in the United States alone, and they admit it is a conservative estimate—more realistic estimates are over 100,000 people a year.<sup>7</sup> To give some sense of proportion, CDI in the United States is far bigger in terms of mortality than AIDS.

I did try to treat our first patient for another seven months with more antibiotics. As I was becoming convinced that antibiotics were not going to offer a cure, I discussed the ethics of doing a fecal infusion with my regulatory colleagues at the University who agreed that we had to try this treatment. However, I thought it was critical at that point to try and understand what actually happens during this procedure. Although clinicians as far back as Dr Eiseman believed that this approach should reintroduce normal bacteria back into the intestinal tract and repair its disrupted microbial ecology, the scientific paradigm at the time held that gut microbial communities were extremely resilient and unalterable. The obvious questions were whether the procedure would lead to engraftment? How much? Would any changes be sustained? Technologically, these questions were ripe for some experimental testing. However, I could not possibly do it by myself because I was not trained as a microbial ecologist. So, I decided that we needed to form a team, and I looked for a microbial ecology colleague at the University. That is how I met Dr. Michael Sadowsky, head of the BioTechnology Institute and a highly accomplished microbial ecologist. The team approach became fundamental to everything that followed.

After reviewing the sparse literature on what was then called “fecal bacteriotherapy,” I decided that the safest and surest way to introduce the material was by way of a colonoscopy. I prepared a crude suspension of her husband’s fecal material in normal saline using a kitchen blender. Incredibly, while still in recovery, the patient said she felt something in her was becoming whole and she was feeling better! After having diarrhea for fifteen months, she had her first normal bowel movement two days after the treatment.

We analyzed the fecal samples taken before and after the procedure with the help of Drs Jansson and Dicksved in Sweden, and demonstrated that our patient now had donor-like bacterial composition in her colon. Furthermore, engraftment of the donor microbial community was sustained over a month. This was the first demonstration that it is possible to transplant an entire microbial community in an adult human.

**Mr LeBeau:** That case became big news in the medical world and among the general public.

**Dr Khoruts:** The results were published in the *Journal of Clinical Gastroenterology*,<sup>8</sup> and the story was picked up by *The New York Times*. The case quickly became a poster child for this newly emerging area of science

because there was a clear-cut translational element, without which it would seem too abstract and complex to be applicable to anything other than science fiction.

I think many scientists found the results to be inspiring for a variety of projects in microbiota research because the clinical relevance became immediate. Since then, there has been an exponential growth in this whole new scientific frontier. The results also provided a scientific foundation for this treatment, and many physicians started developing their own fecal transplantation programs. I also started treating more and more patients with CDI. The first case I ran into was certainly not an isolated problem, and we continued to build a clinical program at the University of Minnesota dedicated to treatment of recurrent CDI. At this point, we’ve done approximately 200 patients with recurrent CDI over 4 or 5 years. Our treatments have a 90% efficacy with one instillation.<sup>9</sup> If patients fail the initial treatment and choose to have it done again, they still have a 90% chance of succeeding the second time. That brings the overall success rate to approximately 99%.

**Mr LeBeau:** Have you made any changes in your process as you developed your clinical program?

**Dr Khoruts:** I did the first 10 transplants the old-fashioned way with a blender in the endoscopy bathroom. During that experience I quickly appreciated the practical barriers to doing fecal transplantation in a busy clinical setting. The olfactory potency of human fecal material revealed at the touch of a button on the blender can be quite shocking—it can empty the waiting rooms. Importantly, donor identification and testing can be a big challenge, especially as these efforts are largely unreimbursed. Even procurement of fresh material can become a logistical problem.

We introduced a number of innovations. We developed a formal volunteer donor program. It is cruel to charge sick patients with finding donors, and people they were able to bring were typically not necessarily healthy. Given the importance of microbiota in human physiology and our ignorance about most of it, I felt uneasy making compromises on donor health. We also developed a standardized methodology in preparing the microbial fraction from the fecal material and learned to cryopreserve it.<sup>9</sup> These innovations largely addressed the practical challenges associated with crude fecal transplantation.

At this time, securely collected fecal material is transported to a laboratory on the St Paul University campus, which is an FDA-registered site for Good Manufacturing Practices (GMP) production of molecular and cellular therapeutic materials. GMP manufacturing ensures that the product is manufactured according to strict protocols. All steps are documented and ready for inspection at any time. While we cannot guarantee that the composition of the material is the same for every lot, we can guarantee that every lot was

produced the same way. As fecal microbiota transplantation continues to move into mainstream medicine, we feel these details become absolutely critical.

**Mr LeBeau:** How can you be sure that the freezing process doesn't kill off any of the good bacteria?

**Dr Khoruts:** We follow the composition of fecal microbes in the recipients before and after the fecal transplant.

**Mr LeBeau:** By looking at their genetic sequence?

**Dr Khoruts:** Most of bacteria cannot be cultured outside the gut using classical microbiological techniques; one cannot test viability by growth in a Petri dish. We check for whether the bacteria are dead or alive by indirect assays such as measuring cell membrane integrity. But the real test is what happens in the recipient. Do these bacteria start proliferating? Do they become this person's new microbial community? Studying the fecal microbiome of recipients is the ultimate and most relevant test, and it remains central to our work.

**Mr LeBeau:** Why can't the bacteria grow outside the gut?

**Dr Khoruts:** These microorganisms live in a community that we cannot reproduce in a Petri dish. They are social, just like humans. You can imagine putting a person in an isolation cell with no stimulation. This person is going to become depressed and ill and die sooner than he or she would in a normal, nurturing, interactive community. It is similar for individuals within a healthy microbial community. Individual microorganisms are highly specialized in their functions, and they are interdependent. One microbe may do a particular part of a chemical reaction, but it is also dependent on what a neighbor does, etc.

When researchers try to grow these bacteria using classical microbiological techniques, one organism at a time, they end up missing 99% of species. In fact, microbes that can be grown easily in the laboratory as single organisms are often pathogens. If you ask most physicians or medical students to name a fecal microorganism, they typically name *E coli*. Many strains of *E coli* are also notorious for causing diarrheal disease. In reality, even non-pathogenic *E coli* is a very minor member of normal microbial community in the colon—it just grows well in the lab.

Until the last decade or two, we had no clear idea of what composed the gut microbiota. Metagenetic and computational technologies that emerged only recently and continue to evolve rapidly enabled us to ask this question. Over the last couple years several major research initiatives, including the NIH-sponsored Human Microbiome Project and the MetaHIT project financed by the European Commission, established the basic framework defining normal microbial communities in the gut and various other sites in the human body. This work is very much in progress, as the full diversity of human experience has not yet been captured.

**Mr LeBeau:** What are the most immediate challenges facing your program?

**Dr Khoruts:** One major challenge is recruitment and retention of donors. As I noted earlier, we felt that tasking ill recipients with finding their own donors was impractical, suboptimal in terms of safety, and frankly, cruel. We embarked on building up a community of healthy volunteer donors. It turns out there are relatively few truly healthy people. We end up disqualifying approximately 90% to 95% if people we're able to recruit. All these individuals pass rigorous infectious disease testing. However, problems of food intolerances, obesity, metabolic abnormalities, autoimmunity, allergies, asthma, and neurologic and psychiatric disorders are very much commonplace. Current science tells us that microbiota may be contributing to all these conditions. Therefore, they become exclusion criteria in our program.

We are now perpetually recruiting new donors. However, donation of fecal material is a new paradigm in the transplant world. Many people feel uncomfortable with the idea or find it humorous. This is even true among most healthcare workers and students. Recently a medical student came through the initial screening examination, responding to a recruitment flyer. She mentioned her intent to participate to her friends, other medical students. They all laughed. Of course, nobody would laugh at a blood donor. In fact, our program is unmatched in its potential to save lives. One simple donation can help save several people from dying or having a miserable existence. No blood donation can do that. We have a lot of educational work to do.

**Mr LeBeau:** Is fecal transplantation regulated by the US Food and Drug Administration (FDA)?

**Dr Khoruts:** The FDA seemed to look the other way until last year. In May of 2013, the FDA organized a public workshop. They invited researchers of the gut microbiome as well as a number of clinical practitioners of FMT. At the end of the workshop, they presented their own viewpoint—FMT is a “drug.” They explained that Congress has defined any product that claims to mitigate, treat, cure, or prevent disease to be a drug. Products made from human cells and tissues fall under a different classification (Human Cells, Tissues, and Cellular and Tissue-based Products, or HCT/Ps). However, the FDA has determined that fecal microbiota are not *human*. Scientifically, I think they are wrong about this point. Microbiota have co-evolved with their host, and composition of microbiota is species-specific. In fact, nobody has raised the possibility of using dogs, cats, or mice as donors of fecal microbiota for human patients. However it may be, the precise classification is merely a regulatory wrinkle. Both drugs and HCT/Ps are regulated by the FDA.

However, if FMT is a drug (in the category of biologics), it is currently an unapproved drug. The FDA requires all practitioners using an unapproved drug to submit an Investigational New Drug Application (IND). At the

workshop, the FDA informed the community that an IND is expected to be filed by any clinician administering an FMT. This announcement made headlines everywhere and was perceived as a heavy-handed governmental overreach. Indeed, by that point the evidence was very convincing that FMT is lifesaving for people suffering from refractory CDI, including a randomized, controlled study from Europe. However, the burden of filing an IND made it essentially unavailable to physicians. An IND is a challenging application—our team worked for a year to put it together. At the end, it weighed 22 pounds and had to be packed in multiple boxes to be mailed to the FDA! This amount of paperwork is routinely handled by pharmaceutical companies, and they have dedicated full-time staff just for regulatory paperwork. However, individual physicians can't possibly be expected to do something like this. So, even though the FDA did not intend to simply shut down FMTs, their normal operating procedures had that effect on the ground.

There was an outcry of protest from the public and physicians. Stories in newspapers featured desperate patients already scheduled to get the treatment only to be told by their providers that the procedure was canceled. I started getting more emails from patients from all over the US wishing to fly out to Minneapolis for the procedure. Various physician groups wrote to me asking for a copy of our IND or to be included as a site under our program. Naively, they did not understand that an investigator holding the IND becomes responsible for everything done under its umbrella. We simply didn't have those kinds of resources. Ultimately, several physicians including myself, who did get an IND granted by the FDA, published a guide to physicians for how to write this application.<sup>10</sup> Frankly, I am not even sure how the FDA planned to police all these INDs—there were potentially hundreds of individual physicians and groups they would need to monitor. Indeed, in July of 2013, the FDA published an amendment to their initial statement, which declared they will exercise “enforcement discretion” and allow administration of FMTs without an IND for CDI that is not responding to standard antibiotic treatments; however, all other indications still require an IND. In essence, the FDA declared a “Don't ask, don't tell” policy regarding use of FMTs in treatment of CDI.

**Mr LeBeau:** How long is the FDA going to exercise this enforcement discretion?

**Dr Khoruts:** They could pull that back at any time. No one knows for sure. I actually do feel that the FDA has a responsibility to ensure a thorough drug approval process for FMTs. There are many potential dangers, and desperate patients can easily become victims. Some predatory practices have emerged already. I learned from one person looking for an FMT that a local practitioner in her area charged \$10,000 per treatment. I am even more concerned that some organizations or companies can use the current regulatory paralysis and embark on mass manufacturing of fecal microbiota

material without having to adhere to strict protocols. This appears to be happening already. The FDA has demonstrated over the course of last year that they can act flexibly and swiftly. It is critical that they don't abdicate their responsibility for ensuring the safety and well-being of current and future patients.

**Mr LeBeau:** There's also a movement to promote do-it-yourself fecal transplants. There are websites and even books promoting at-home procedures. What do you think of the idea of do-it-yourselfers?

**Dr Khoruts:** I get emails all the time from people who want to take somebody's feces, put them in a blender and do an enema. In my experience, people who choose to perform do-it-yourself FMT generally do not do it for CDI. They do it for other indications, such as inflammatory bowel disease, irritable bowel syndrome, autoimmunity, and many other problems for which medicine is not offering options or medications have many potential or real problems. I don't think the do-it-yourself experiments are happening on a large scale at this time, but there is a serious danger that unregulated mass manufacturing will find a market.

**Mr LeBeau:** Do you advise people against do-it-yourself FMT?

**Dr Khoruts:** I don't. I hear from people who have read on the Internet stories of how their inflammatory bowel disease (IBD) gets cured with a session or two. For example, *The New York Times* published a commentary by a donor to a friend of hers, “Why I Donated My Stool.” She described how she made a donation and cured her friend. People read those kinds of stories; they also read that fecal transplants have no risks or side effects, and they want to give it a try.

In fact, there are no systematic trials of any reasonable size. All current evidence for non-CDI indications is anecdotal. If one looks at IBD trials of other drugs, there is a 30% placebo response rate. This disease is characterized by spontaneous relapses and remissions. We do not know what causes flaring of disease activity or what causes the remissions to happen on their own. In IBS the placebo response rate is even higher. Obviously people who have a good outcome are more likely to share their experience. People will be less likely to talk about an activity that may be pseudo-illegal. They are even more likely to feel stupid to admit what they did if it didn't work. The potential for ridicule is great. After all, they're putting someone else's poop in themselves thinking it will cure their disease?

In fact, the early systematic data are mixed. Two tiny studies were recently reported from Austria on using FMT in treatment of severe ulcerative colitis.<sup>11,12</sup> Both showed essentially no clinical benefit beyond what might expect above placebo (these were not placebo-controlled). In fact, some patients suffered flares of their IBD activity. Yet there was a hint that some benefit



could be observed if there was rise in the abundance of certain bacteria thought to play beneficial roles.

**Mr LeBeau:** Why can an FMT be so effective for CDI but more uncertain for things like ulcerative colitis?

**Dr Khoruts:** The microbial community in the gut of patients with recurrent CDI has been devastated by antibiotics. Our average patient has been carpet bombed with antibiotics for a year! When we study the residual microbiota, it looks nothing close to what is normally seen in the gut. The microbial diversity is markedly reduced, and residual microbial species that become dominant are not even normally common. When a healthy microbiota is introduced, there is prompt restoration of normal gut microbial composition and function.

However, ulcerative colitis and many other diseases are very different. In all these conditions there is an established microbial community. It may be somewhat altered, but we don't know whether the differences are the cause or the effect of the disease. The microbial diversity may be slightly decreased, but that drop is nothing like what is seen in recurrent CDI. Merely introducing a whole bunch of new microbes does not necessarily result in creation of an entirely new microbial community. If you think about it, what do farmers do? Before they put seeds in the ground, they plow the field. They prepare the field, right? If you are just going to go into the woods and throw seeds around, you may get some sprouts, but you're not going to get much of a crop. You need to clear the land, you have to plow the ground, and then you can put in the seeds. It is the same here. First you have to remove the old community. That likely will require an antibiotic conditioning regimen, which is yet to be established.

**Mr LeBeau:** So you could deliberately decimate one's gut microbiota in order to plant a new crop, so to speak?

**Dr Khoruts:** Yes, but you have to have some thought into this and systematically try out different antibiotic regimens. It is important to note that residual antibiotics can also impede engraftment. However, I am confident these important details can be sorted out. The next question is who is the appropriate donor. It is possible that virtually every individual raised in the United States already has altered microbiota caused by altered diet rich in processed foods and significant exposure to antibiotics. Just because the person does not have ulcerative colitis does not mean the person's microbiota does not have the potential to cause the disease in a genetically susceptible individual. We know that microbiota composition in people living in ancestral communities in Africa and South American Amazon forests has significantly higher microbial diversity and functional potential for digestion of complex polysaccharides.<sup>13</sup> Are those microbiota more likely to be therapeutic? Or, perhaps, our microbiota are already adapted to the new environmental pressures of living in the developed world, and trying to recreate ancestral microbiota may make some diseases even worse.

**Mr LeBeau:** Some of the do-it-yourselfers that I've read about online say they were CDI patients who went to their physicians and were turned away because their doctors found the whole process to be disgusting. They were subdued by the "yuck factor."

**Dr Khoruts:** You are correct. The doctors say "yuck" more than the patients, but probably because these doctors envision a very crude version of it, which is already in the past.

**Mr LeBeau:** They don't want to go into the back room with a blender?

**Dr Khoruts:** That's true. It is disgusting and it is probably not a good idea. General (Occupational Safety and Health Administration) laboratory guidelines require a biological safety cabinet when working with human feces. Although donor fecal material in this case comes from truly healthy individuals, I don't think aerosolized fecal material is necessarily safe. Nature did not envision breathing aerosolized fecal particles. I can imagine one could stir up some inflammation in the lungs. That is actually a controversial point. The FDA says nothing on the topic, and people interpret that as okay. But it is not the FDA's job to worry about the physician; its job is to worry about the patient.

**Mr LeBeau:** This means there's a market out there for pre-processed fecal material, like the frozen doses that you produce.

**Dr Khoruts:** We largely solved the aesthetic problems, and we completely solved the burden of donor recruitment, screening, and testing. The material hardly smells. It has a brown color still because various chemical pigments have not been completely removed. It is just a suspension you can put in a syringe and deliver into a patient during an endoscopic procedure like colonoscopy.

**Mr LeBeau:** Are you in a position to start marketing at the University of Minnesota?

**Dr Khoruts:** I get inquiries about this frequently. There is a demand. Unfortunately, the FDA has not given a clear answer regarding whether this would be legal, even at mere cost of production. Currently, we prioritize production for various clinical trials. However, not everyone is so patient.

**Mr LeBeau:** What do you mean?

**Dr Khoruts:** A group of students at the Massachusetts Institute of Technology are explicitly using the protocols we developed and published on preparation and cryopreservation of fecal microbiota and set up an organization or a company called "Open Biome." They are selling this material nationwide for \$250 per dose. This group has interpreted the July 2013 FDA statement to

mean that fecal microbiota can be mass-produced and marketed for treatment of CDI.

**Mr LeBeau:** What does the FDA say about Open Biome?

**Dr Khoruts:** They are not saying anything. The FDA appears to be paralyzed. However, I do hope it articulates what the rules are. I am personally somewhat torn on the issue. Whatever Open Biome is doing is probably superior to what physicians may be doing on their own. However, there are also clear dangers.

**Mr LeBeau:** Does OpenBiome have an IND?

**Dr Khoruts:** Yes. However, that does not mean that they received a permission from the FDA to market this material nationwide. I asked the FDA several times to tell us if we can sell the material at cost of production to other groups. Their answer was that this is an issue being actively discussed at the FDA. They've given the same answer for months now.

**Mr LeBeau:** What are the dangers, and what do you want the FDA to do?

**Dr Khoruts:** Short-circuiting the approval process for a drug is never a good idea. I do think there is a need for an accelerated pathway toward approval. However, once there is a generic version of a pharmaceutical product already on the market, all research and development typically stops. Patient recruitment into trials becomes impossible. We don't often see studies on old drugs that are now generic, even though we still have a ton to learn. Unfortunately, the government does not sponsor the needed clinical research, expecting that to be done by pharmaceutical companies. However, pharmaceutical companies are not likely to invest in the development of a product that they will not likely be able to monetize some day.

I would be very surprised if the FDA would allow mass production and sale of a biologic drug that may not be manufactured by GMP protocols. A biologic therapeutic is defined by *the process* of manufacturing since the finished product is always variable in precise composition. There is always the potential that some manufacturers will try to cut corners somewhere, which cannot be good for the public. Some may brush these concerns aside—after all, this is merely natural human microbiota. That is my point, exactly, however. This is highly complex biologic material with significant potential to do benefit or to do harm. The FDA was right to step in last year, even though their execution was clumsy. They continue to have a responsibility now.

Ultimately, the main thing I'd like from the FDA is clarity. At the very minimum, all manufacturers should play by the same rules. The public is best served when there is meaningful regulation and robust competition.

**Mr LeBeau:** If the FDA takes your advice and says, okay,

we will regulate it and you could sell it at cost, would the University start doing it?

**Dr Khoruts:** We would.

**Mr LeBeau:** Are there any other business models that might crop up in the *C difficile* market?

**Dr Khoruts:** There are various ideas. The whole microbiome area is something that has attracted pharmaceutical interest. The Big Pharma are exploring ways to get involved. However, they are not quite jumping into the whole fecal transplant thing yet. One major concern is that the intellectual property landscape is unclear, and that scares the business people. However, there is also the idea that good service and a safe product can be a good business model.

There is also great interest in developing synthetic mixtures of microbiota. This clearly addresses some of the intellectual property concerns, but the approach is not without challenges.

**Mr LeBeau:** What are the pros and cons of synthetic products?

**Dr Khoruts:** The challenges associated with putting together synthetic mixtures should not be underestimated. For decades, researchers and companies have been trying to manufacture a synthetic blood replacement. However, it has been very difficult to create an artificial oxygen-carrying alternative to blood, and thus far all different synthetic versions have failed in clinical trials. I think we are a long way from confidently putting together synthetic microbiota that would equal something made by nature over many millions of years.

A Canadian team of researchers recently developed a synthetic mix they call RePOOPulate, grown in a bioreactor called "Robogut," and successfully treated a couple patients with CDI.<sup>14</sup> However, their choice of microorganisms was not based on mechanistic understanding. Instead, it was based on the ability to grow bacteria outside the human body, antibiotic sensitivity, and associations of individual organisms with specific infections or diseases. A truly mechanistic approach would be based on knowing what functions the microbial community needs to perform, eg, produce targeted anti-*C difficile* compounds, stimulate protective immune responses, or something else. We and others, for example, suspect a critical role for bacteria that transform primary bile salts into secondary bile acids in the colon in control of CDI. In addition, one needs to understand factors that make microbial communities stable and resilient. The fact is that patients with recurrent CDI are often exposed to antibiotics for non-CDI indications. Each new antibiotic exposure is like a hurricane to a microbial community. A resilient community is able to recover, which is why antibiotics are generally well tolerated by most people.

Health Canada at the moment has put RePOOPulate

on hold. They want more research and data, such as complete DNA sequencing of all the microorganisms in the mixture. I don't think that is the most relevant request. I think these data can just make someone feel better that there is "a lot of research." The relevant questions should be whether the microbial community that was put together is stable at least in the bioreactor—making sure that every lot is consistent. Furthermore, the fact that the mixture may look taxonomically identical doesn't mean that every lot remains functionally the same. The bioreactor isn't the human gut, and one can expect the microbial community to evolve and adapt to conditions within the bioreactor. This can lead to loss of physiologically important activities in favor of something that helps life in the reactor.

Proponents of defined mixtures often say they offer a lower risk of infection. However, I don't think the evidence supports this concern so far. Risk of infection associated with fecal transplants appears to be extremely low. Furthermore, in a transplant model a single donor can only ever do limited harm even if some infection was missed in donor testing. However, a contaminated bioreactor can potentially infect thousands of people. Contamination is a real concern in my mind—I trust the immune system of the donor far more than a technician in a manufacturing facility. Furthermore, we should not ignore potential long-term problems. How do we know that some mixture put together by humans with very limited knowledge isn't going to lead to colon cancer or some other disease in 10 years?

**Mr LeBeau:** Are the synthetics easier to regulate?

**Dr Khoruts:** Regulatory agencies are seduced by the idea of defined mixtures. It gives them a target for regulation. It is easy to demand compositional consistency. That may be important, but there are many other important factors to consider. The fact is that fecal transplantation offers a full microbial community designed by nature and tested in the original donor for decades of that person's life. That is a very high bar to reach.

The regulatory bodies are faced with new scientific paradigms. For decades, the focus has been on individual pathogens while commensal host microbial communities were completely ignored. Even now, clinical trials of antibiotics consider short-term endpoints only. Today, we are legitimately concerned about potential contributory effects of antibiotics on many diseases that have become so prevalent in our society, including diabetes, autoimmunity, allergic diseases, inflammatory bowel disease, psychiatric disorders, autism, and many others.<sup>15</sup> The regulatory bodies also need to educate themselves in the discipline of microbial ecology and the emerging science of microbe-host interactions. They need to learn key concepts such as stability and resilience of a microbial community, and we still need to learn reliable indicators that can predict these properties. They need to understand that microbial communities are intrinsi-

cally dynamic—precise taxonomic compositions change constantly depending on specific meals and activities of the host. There has to be scientific understanding on what constitutes variation consistent with this dynamic behavior versus pathologic deviations. This requires focus on functional assays of microbiota.

Obviously, manufacturing and scientific challenges should be associated with corresponding rigor of regulation.

**Mr LeBeau:** You described a lot of skepticism about synthetic microbiota. Are there any positives?

**Dr Khoruts:** Absolutely. The approach has enormous potential for scientific progress in this field. By trying to create synthetic microbial communities, we are going to learn a lot about how microbial communities are put together. We are going to learn more about likely critical contributions of the host immune system in shepherding these microbial communities. Furthermore, as we aim to develop various synthetic mixes, we are forced to think about mechanisms. A particular property that may be essential to control *C difficile* infection may be irrelevant in ulcerative colitis. However, in ulcerative colitis we may strive for a microbial community that calms down gut inflammation. We may wish for something still different in autism, etc. Yes, the synthetic approach is very important for advancement of science and ultimately may lead to emergence of very important new therapeutics. I am ultimately optimistic and very supportive of this work.

**Mr LeBeau:** Do probiotics fit into treatments of *C difficile* or any of the other diseases? They are supposed to improve the microbial communities in the gut.

**Dr Khoruts:** It is critical to understand that probiotics by definition are never intended to treat any specific diseases. Scientifically defined, probiotics are live microorganisms that benefit human health. Although that sounds good, I am actually not sure what that means and I don't think anyone else is either. We don't have an accepted definition of "health" that is not merely absence of disease. There is also no legal definition of probiotics, and manufacturers have to be very careful not to claim an indication for disease treatment—that gets them into the "drug" category and invites a rigorous regulatory burden of proof in efficacy.

Obviously, probiotics constitute a multibillion dollar industry. However, it is built mostly on creative marketing rather than clinical science. The manufacturers make nebulous claims like "balancing your flora." Well, in reality, we are only now beginning to learn what normal and healthy microbiota actually looks like and there is no scientific agreement on that yet. Virtually all my patients take probiotics for their *C difficile* infection. There is very little chance or evidence that it is doing them any good. Yet I don't stand in the way of that because patients need to feel that they have some con-

control over their situation. I do express my uncertainty about the benefit so they can make an informed decision on whether they want to spend the money.

**Mr LeBeau:** Does the FDA regulate probiotics?

**Dr Khoruts:** Yes, but I think the FDA has done a very poor job of regulating probiotics. There is no special definition for probiotics used by the FDA, and products are regulated based on whether they fall into one of the existing regulated product categories, eg, drugs, biologics, foods.<sup>16</sup> While most microbial species and strains used as probiotics are likely harmless, it is very unlikely they actually do anything of importance in the human digestive tract. The organisms were never chosen based on understanding of microbe-host interactions, microbial ecology principles, or anything to do with human physiology. Some emerged merely as microorganisms that ferment dairy products and other foods and became associated with healthy living by cultural heritage. Thus, advanced age is highly respected in some southern cultures, which also had historically relied on fermentation rather than refrigeration to preserve foods.

Even the most basic questions such as actual counts of organisms are controversial. Researchers working with probiotics know that specific vehicles for these products, eg, encapsulation, dairy products, etc, constitute critical variables for viability of these microorganisms both outside and inside the human host.<sup>17</sup> Even one single probiotic strain isolated from different specific products was shown to have vastly different properties in specific tests that presumably defined their beneficial properties.<sup>18</sup> By all indications, regulation of probiotics by the FDA today is extremely lax. I am guessing that this entire area is not considered a high priority for them because probiotics do not generally want to enter the drug category. There also is likely massive industry pressure to leave things as they are, maximally ambiguous. Marketing is working well and the demand is high. Why spoil a good thing with some science?

**Mr LeBeau:** What do you expect and hope will happen in the area of microbial therapeutics over the next decade?

**Dr Khoruts:** First, I like to define this category as “microbiota” rather than “microbial” therapeutics. That gets us away from the infectious disease connotations typically associated with the word “microbial.” It recognizes entire microbial communities as distinct entities rather than mere sums of individual microorganisms. Finally, the category is explicitly meant to treat disease, which sets it clearly apart from probiotics.

I do expect emergence of an entirely new class of drugs called “microbiota therapeutics.” Fecal transplants clearly represent one member of this class. I hope the regulatory agencies get updated on the current sciences involved in this field and aren’t guided by outdated concepts and inappropriate concerns. I do believe that the regulatory agencies have a critical role

to play in nurturing this area of development. If executed correctly, we will have new remedies that are safe and highly effective for some important problems that have no adequate solutions today.

Personally, I think the most likely early successes will be in controlling antibiotic-induced complications that can benefit from prompt restoration of normal microbial gut ecology. CDI is one example of that, but there are likely many others. I think science will continue to develop rapidly, and ultimately we will be in a position to design specific microbiota-based therapeutics for other important conditions such as diabetes and inflammatory diseases. However, that goal will likely require more than a decade to become a reality in mainstream medicine.

I suspect that as the field grows, we will see more tension between probiotics and microbiota therapeutics. However, I hope industry will see the great potential of the entire field and scientific development will prevail over clever marketing alone. Currently, the industry is largely sitting on the sidelines of microbiota science or may be engaging at a very low level. But successes will generate genuine support and an accelerated pace of research.

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