C. difficile Infection and Recurrence: Targeting the Drivers of Pathogenesis

Question: Can you please start our discussion on C. difficile infection and its recurrence by describing the body’s defense mechanisms against C. difficile?

Dr. Feuerstadt: Our body has at least two defense mechanisms against C. difficile, including colonization resistance and our serologic or blood-borne immune system response. In order to best understand these mechanisms, let’s quickly review the infection and its mechanism of infectivity. In the environment, C. difficile rests in an indolent, inert state called the spore phase. And classically what happens is we swallow the spore phase, it’s resistant to our gastric acid, it gets into our small bowel where it converts or germinates into the vegetative phase. The vegetative phase multiplies, divides, and multiplies as it builds steam heading towards the colon.

But the colon is a very smart organ. The colon has two independent mechanisms to prevent C. difficile proliferation. One of those defense mechanisms is the bacterial milieu; the so-called colonization resistance. What classically weakens colonization resistance? Amoxicillin, ampicillin, clarithromycin, fluoroquinolones and cephalosporins. When the colonization resistance is weakened, the C. difficile is much more likely to proliferate and take over.

Within the vegetative phase there are two main toxins that are released: toxin A and toxin B. And these toxins alter the epithelial layer of the colon, resulting in weepage of fluid across the bowel, pseudomembranes and the diarrheal state most commonly associated with C. difficile. The blood-borne immune system, when the epithelium of the colon is compromised, can actually release immunoglobulins into the lumen of the bowel that bind toxin A and toxin B in specific ways, and that can control the infection and minimize the infection from taking over. So, between colonization resistance and our immune system
response, there are two mechanisms that our body can protect against *C. difficile*.

**Question:** Since restoration of the gut microbiome is an important aspect of treating *C. difficile* and recurrent disease, can you please provide an overview of the microbiome shift that occurs during and after *C. difficile* infection?

**Dr. Feuerstadt:** There are microbiome shifts that occur prior to the infection and throughout that are important to understand in order to best treat patients with *C. difficile*. The three shifts that patients experience and the microbiota experiences are termed microbiota suppression, collateral damage and the window of vulnerability. Let’s first focus on the microbiota suppression.

And this is what we just discussed in terms of antimicrobials weakening colonization resistance. The best study that looked into this was a study by Chang et al. Chang looked at ten total patients. Three individuals had no *C. difficile* infection. Four individuals had an initial infection, and three individuals had recurrent *C. difficile*. Chang compared the diversity of the microbiome as well as the constituency between these three groups (Figure 1). When the group that had no *C. difficile* was compared with the group that had initial infection, the group that had initial infection had a depletion of the Bacteroides and the Firmicutes, but there was no difference in the diversity. So it is believed that the Bacteroides and the Firmicutes play an essential role in prevention of the onset of *C. difficile* and play an important role in colonization resistance.

However, when the group with initial infection was compared with the group with recurrent infection, the group with recurrent infection had two hits. They had depletion of Bacteroides and Firmicutes, but they also had a depletion of diversity. And it’s believed that the depletion of diversity combined with the alteration of the Bacteroides and the Firmicutes results in recurrence after
recurrence that we see in the quoted 40 to 60% recurrence rates moving forward.

The second phase of alterations to the microbiota that we see is something called collateral damage. And collateral damage is the concept that although we have antimicrobials that are effective at treating *C. difficile*, they also alter the microbiome. And by altering the microbiome they create an environment where colonization resistance is weakened, in addition to weakening the *C. difficile*, and therefore patients are left at risk for recurrence. And this is believed to be why about 25% of patients that receive either metronidazole or vancomycin will recur in the future.

And further, the third concept is something called the window of vulnerability. The window of vulnerability is the timeframe from completion of therapy to three months after completion of therapy when patients are most vulnerable to a recurrence of this infection. Probably the best study that looked into this was a study by Drekonja et al. Drekonja looked at 246 individuals, all of which had *C. difficile* infection. But Drekonja separated the patients into three groups. One group that received no other antimicrobials for independent indications, one group that received antimicrobials for other indications at the same time as their treatment of *C. difficile*, and a third group who received antimicrobials for other indications in the three months subsequent to the treatment of the *C. difficile*.

Interestingly enough, when they looked at the recurrence rates, recurrence rates in the group that received no antimicrobials was 16%. Recurrence rates in the group that received antimicrobials at the same time as their treatment of *C. difficile*, shot up to 31%. However, recurrence rates in the group that received antimicrobials in the three months subsequent to the treatment of *C. difficile*, within the window of vulnerability, shot up to 48%.
So, the bigger question is why? And the answer to that question comes from the concept that antimicrobials suppress *C. difficile* but don’t necessarily completely wipe it out. Plainly stated, at the end of an antimicrobial course an effectively treated patient and a patient that potentially is cured still has *C. difficile* lurking in their system. What happens next is that the colonization resistance regrows and the colonization resistance provides the knockout punch. As was evidenced in the Drekonja study, when the colonization resistance takes a hit during this subsequent three-month time period, that weakens it and recurrence is much, much more likely.

We can see that by understanding these three alterations to the microbiome, we can better understand *C. difficile* infection, but also provide a framework for better understanding of more efficient therapies and therapeutics for *C. difficile*.

Question: Can you describe the immune system response to *C. difficile*?

Dr. Feuerstadt: Yes, now that we have discussed the microbiome shift associated with this infection, let’s focus on the serologic or blood-borne immune system response to *C. difficile*. And the original ideas behind this came from Lorraine Kyne and Ciaran Kelly back in the late 1990s. They did something very, very smart. They looked at a cohort of patients in their healthcare system between January of 1998 and May of 1998 who were admitted receiving antimicrobials for any indication. Overall, they included 271 individuals and they checked their bloodwork and their stool studies every third day for three months after the initiation of the other antimicrobial. Of those 271 patients, 47 became positive for *C. difficile*. However, 19 of them never developed the infection.

What separated those 19 who were asymptomatic but positive for *C. difficile* from the remainder who were positive and had active infection? What differentiated them was an immunoglobulin response to toxin A, an IgG
response to toxin A. In fact, the group that had an IgG response to toxin A was 48-fold less likely to come down with symptomatic *C. difficile*. So, it seemed as though this immunoglobulin response was suppressing the infection and the infection’s ability to act on the lining of the bowel.

They subsequently did something even more interesting. They looked at a cohort of 63 patients who had active *C. difficile* and were being treated. They checked their bloodwork and stool studies every third day for three months after the initiation of treatment of *C. difficile*. What they found was that 22 total patients had recurrent disease, but what differentiated those who recurred from those who did not recur was an immunoglobulin response to toxin A again (Figure 2). In fact, within three days the group that did not recur had a statistically significantly higher IgM response to toxin A. And within 12 days and beyond the group that did not recur had a statistically significantly higher IgG response to toxin A. As a result, this became a therapeutic target that ultimately took the form of a product called bezlotoxumab, which is a fully humanized, monoclonal antibody to toxin B indicated to prevent recurrence of *C. difficile* infection.

So, we can see that by understanding the immune system response to *C. difficile* and the basic premise, a therapeutic target was designed and now can be implemented.

**Question:** What impact do available antimicrobial therapies for treating *C. difficile* infection have on the gut microbiome?

**Dr. Feuerstadt:** Fidaxomicin and ridinilazole are examples of antimicrobial therapies with less effects on the gut microbiome. Fidaxomicin is an FDA-approved treatment for initial infection with *C. difficile*, whereas ridinilazole is currently in phase III trials with phase II data presented in October of 2018 at a major infectious
disease conference in San Francisco. These data showed excellent safety and efficacy. However, since ridinilazole data is not published in manuscript form, we will focus on the fidaxomicin treatment as one example of a narrow spectrum antimicrobial that should hypothetically reduce rates of recurrence of *C. difficile*.

In order to best understand the impact of these antimicrobials, one study analyzed the stool from 23 patients with mild to moderate *C. difficile* infection, comparing the impact of ten days of fidaxomicin or vancomycin on the overall microbiome. As hypothesized, fidaxomicin had minimal impact on the phylogenic clusters within the microbiome, maintaining low levels of Enterobacteriaceae and higher levels of Clostridiales. Plainly stated, colonization resistance was preserved. Whereas vancomycin had a profound and statistically significant effect on these essential elements to colonization resistance.

This study by Tannock et al reinforces the concept that narrow spectrum antimicrobials should theoretically minimize the risks for recurrence by fortifying the colonization resistance and improving the sustainability of the initial response to the antibiotic. In the case of fidaxomicin, this is in fact seen, as evidenced by two large phase III randomized control trials, where patients with either initial *C. difficile* infection or first recurrence received fidaxomicin 200 mg twice daily for ten days or vancomycin 125 mg four times daily for ten days. They were then followed for 28 days thereafter for recurrence.

Overall, 542 received fidaxomicin and 563 vancomycin. The initial treatment response was similar between the two groups, with an 88% response rate for fidaxomicin and 87% for vancomycin. What differentiated these two cohorts were their recurrence rates, and one secondary endpoint that accounted for these recurrence rates is something called sustained clinical response. This is a
combination of initial treatment response, which was similar between the two study groups as just discussed, mortality, which was six percent in both study groups, and recurrence, which differed significantly. The sustained clinical response in the fidaxomicin group was 70% in one trial and 72% in the other. Whereas the sustained clinical response in vancomycin was 57% in both trials. This statistically significant difference is believed to be largely driven by the wider impact on the microbiome that vancomycin has compared with fidaxomicin.

It is of course always important to review and understand the phase III clinical trials, but as clinicians, the question that always arises is how effective is a treatment like fidaxomicin in the real world? Do we see decreases in the rates of recurrence for fidaxomicin in the real world? And these questions were considered in a nicely performed retrospective study from England where Goldenberg et al looked at multiple ways that fidaxomicin was implemented into healthcare systems.

The study compared rates of recurrence for those with *C. difficile* infection the year prior to the use of fidaxomicin with the year subsequent to the implementation of fidaxomicin (Figure 3). In two centers where fidaxomicin was used first-line for initial infection, rates of recurrence decreased from 10.6 to 3.1% and 16.3 to 3.1%. In another center where vancomycin continued to be used for initial infection but fidaxomicin was used for first recurrence, overall recurrence rates were decreased from 21.1% to 12.5%. So, we can see that in clinical practice and in the real world the usage of fidaxomicin, a narrow spectrum antimicrobial minimizing collateral damage, seems to reduce rates of recurrence, most likely via preservation of the existing colonization resistance.
Question: A vancomycin taper is often used for the treatment of recurrent *C. difficile* infection. What is the efficacy for this therapy and are there any data on the impact of this approach on patients’ gut microbiome?

Dr. Feuerstadt: Vancomycin is one effective therapy for treating *C. difficile* infection; although it is associated with recurrence. The classic treatment course for initial infection is 10 to 14 days. When considering treating recurrent *C. difficile* infection, vancomycin used in a prolonged and/or staggered fashion seems to be effective. For a prolonged vancomycin taper, the dose of vancomycin is slowly decreased over several weeks. The theory behind this is that by slowly decreasing the frequency of the vancomycin one continues to suppress the *C. difficile* while also allowing the microbiome to rejuvenate and replenish with stronger colonization resistance. As an additional mechanism, sometimes following a vancomycin taper a “pulse” might be given. This is a full dose of vancomycin, 125 mg orally four times daily every third day, and it is usually given for 30 days.

The original study that looked into this was conducted by Christina Surawicz in 2002, where she found that the vancomycin taper in patients with multiply recurrent *C. difficile* infection was associated with a recurrence rate of 31.0% and a pulse was associated with a recurrence rate of 14.3%. Based upon this, the vancomycin taper became the treatment of choice for multiply recurrent *C. difficile* infection for an extended period, and several case series reinforced this efficacy from about 2002 until 2017.

In May of 2017 at a major gastrointestinal conference in Chicago, there was a case series of 128 patients presented from the Mayo Clinic with a median of three episodes of *C. difficile*. The median duration of the taper was 50.5 days, and overall recurrence rates associated with a vancomycin taper was 41%. Really not so good. However, when this study group grouped the tapers
according to being greater than six weeks versus less than six weeks, those greater than six weeks had a recurrence rate of 37.0%. And the duration of the taper was statistically significantly associated with outcome.

This year there was a significant advancement forward with our data considering a vancomycin taper as a very nice meta-analysis was presented at a major digestive disease conference in Washington DC in May. This meta-analysis included eight separate studies conducted between 1985 and 2017 showing that overall vancomycin tapers were able to cure multiply recurrent *C. difficile* 72.8% of the time. This became more interesting when there was a subgroup analysis. When using a taper alone, 61.8% were cured. A pulse alone cured 46.8%. However, if a pulse and a taper were combined, then 89.4% of the patients with multiply recurrent *C. difficile* were cured. This, again, speaks to the efficacy of this regimen to treat challenging cases of multiply recurrent *C. difficile* infection.

Of course, like most things in life, success might come with a cost and a vancomycin taper is no different. A study published by Isaac et al in 2017 in the *Journal of Antimicrobial Chemotherapy* considered the microbiome effect of both short- and long-term vancomycin in humans using high throughput sequencing to analyze the microbiome up to 22 weeks after completion of therapy. Interestingly, during therapy most operational taxonomic units were depleted, including those from the Bacteroides. As we remember, Bacteroides was a key element in colonization resistance and it prevented *C. difficile* infection proliferation within the colon. Following cessation, the recovery of the microbiome was highly variable within the study for unclear reasons, with up to 89% of the operational taxonomic units being undetectable at the longest follow-up time. This lack of recovery of the microbiota that was observed within this study could lead to *C. difficile* as well as other pathogens proliferating within the bowel. Given this, it seems that vancomycin might be
effective at eradicating *C. difficile*, but its aftereffects on the microbiome might lead us to be susceptible to future infections.

Question: Recurrence continues to be a significant problem. In those who have had this infection in the past, is it possible to use available antimicrobial therapies prophylactically to prevent future episodes in those at highest risk for recurrence receiving antimicrobials for other indications?

Dr. Feuerstadt: There is a true fear for recurrence in patients who have had *C. difficile* in the past. There was one observational cross-sectional study that was presented by our study group at a major conference in October of 2018 in Philadelphia. Three hundred and fifty patients with either active *C. difficile* or a history of the infection completed an online survey about the impact of *C. difficile* on their quality of life. Interestingly, of the 235 patients that had the infection in the past, 25% of them expressed a fear of return of *C. difficile* in the future, with 46% fearing the future use of antimicrobials that might leave them prone to recurrence. This fear is real and raised frequently by patients seen in the office. They will commonly ask, is there an antibiotic that I can take to prevent *C. difficile* in the future? The answer is somewhat controversial, since the currently available data is from poorly controlled studies.

In the early 1990s, Dr. Johnson’s group looked into a cohort of individuals colonized with *C. difficile*. This is a group that has the bacterium within their system but does not have an active infection. In effect, their colonization resistance minimizes the ability of the *C. difficile* to proliferate. Within this study, those colonized were treated with either metronidazole, vancomycin or placebo, and then they were followed for two months. In those that received vancomycin, 67% remained positive two months later, whereas only 11% of those that received placebo remained positive two months later.
The theory behind this again focuses on the impact of vancomycin on microbiota. Vancomycin is believed to weaken the colonization resistance in a colonized population, and that allows the *C. difficile* pathogen to remain present. The group that received the placebo didn’t have that impact on their microbiota, and therefore the colonization resistance seemed to be more likely to eradicate the *C. difficile* eight weeks later.

Considering this and the potential collateral microbiome effects discussed previously with regards to vancomycin, the question of whether giving an antimicrobial such as vancomycin as a suppressive therapy might be beneficial when given to those with a history of *C. difficile* who subsequently require antibiotics for other indications is commonly raised. So really what we’re framing here are individuals who are cured of *C. difficile*, but in the future require so-called concomitant antimicrobials.

One retrospective study published by van Hise et al in St. Louis considered this topic. They looked at a cohort of 132 patients that received “no suppressive therapy” when they received concomitant antimicrobials. That group was compared with 71 patients that received either vancomycin 125 mg twice daily or 250 mg twice daily for the duration of the other antimicrobial course. Recurrence rates in those that received no suppressive therapy was 26.6%. However, recurrence rates in those that received the vancomycin was 4.2%, a statistically significant difference. Three of the patients that received the vancomycin suppressive therapy recurred, but two of those three received the 250 mg oral twice daily suppressive therapy. From this data, albeit flawed with a retrospective design and a lack of placebo, it seems that suppressive vancomycin might be beneficial in reducing rates of recurrence and that a low dose might be optimal.
This is still a controversial topic given the known collateral damage associated with vancomycin. However, we know that fidaxomicin has less collateral damage to the microbiome, so theoretically this might be a more ideal therapy to be used prophylactically in patients at risk for recurrence of *C. difficile*. Unfortunately, fidaxomicin prophylaxis used in patients with a history of *C. difficile* infection to prevent future infection has not been studied to date. But, fidaxomicin as a prophylactic agent in a high-risk cohort for initial *C. difficile* infection was presented in 2018 in manuscript form by Mullane et al. This study looked at patients undergoing bone marrow transplant, requiring fluoroquinolone prophylaxis. The patients had several risk factors for *C. difficile* infection, including hospitalization, weakening of the immune system and the prophylactic fluoroquinolone.

They were randomized to either receive placebo or a fidaxomicin 200 mg once daily prophylaxis that was started two days prior to conditioning and continued until seven days after neutrophil engraftment or the completion of fluoroquinolone, up to a total of 40 days. *C. difficile*-associated diarrhea occurred in 4.3% of those that received fidaxomicin compared with 10.7% in those that received placebo, a statistically significant difference. Therefore, in this scenario it seems that the fidaxomicin is an effective prophylaxis minimizing the risks for developing infection. Whether this is applicable to patients who have a history of infection and receive antimicrobials for other indications in the future requires further clarification and study.

**Question:** Fecal microbial transplant is clearly another effective approach for treating multiple recurrent *C. difficile* infections. Are there other ways that we can supplement or impact the microbiome to minimize incidence or recurrence?

**Dr. Feuerstadt:** Yes, fecal transplant is a very effective therapy for patients with multiply recurrent *C. difficile* infection to improve clinically. The question of whether
there are other supplements to the microbiome that might enhance colonization resistance or enhance the ability of the microbiome to eradicate foreign pathogens is something that frequently arises. Let’s first discuss probiotics.

The World Health Organization defines these as live microorganisms which, when administered in adequate amounts, confer a health benefit on the host. These take the form of supplements most commonly available over the counter in the form of *Saccharomyces boulardii*, *Lactobacillus*, *Bifidobacterium infantis* as individual elements or some combination of these components with other probiotics. There are a couple of ways that one might consider using probiotics for *C. difficile* infection.

Firstly, would be as a primary prevention when an at-risk patient is undergoing other antimicrobial therapy. The second application would be adding probiotics to the treatment of *C. difficile* and continuing it following treatment to prevent future recurrence. For primary prevention of *C. difficile* in a population at risk for infection, meaning those with primary risk factors, who undergo antimicrobial therapy, probiotics are frequently given to prevent either antibiotic-associated diarrhea and/or *C. difficile* infection.

One meta-analysis published in the *Annals of Internal Medicine* in 2012 considered 20 studies and approximately 3900 patients who received primary prophylaxis with either *Saccharomyces boulardii*, *Bifidobacterium infantis*, or *Lactobacillus* for the prevention of *C. difficile*. Overall, the probiotics were associated with a risk reduction of 66%. And that was very convincing evidence, but again, it is always interesting to see a practical clinical application of this concept. And that was put forward at a major GI conference in Chicago in 2017. At that conference, West Carstensen presented a study where three internal medicine practices started using *Saccharomyces boulardii* prophylactically twice daily when any other antimicrobials were being given
for non-*C. difficile* related diseases. They compared the incidence of antibiotic-associated *C. difficile* infection and found an incidence of 1.3% in those that received the *Saccharomyces boulardii* versus 3.6% in a historical control. So, we can see a statistically significant reduction compared with the historical measures.

The most definitive study to date considering probiotics for prophylaxis for *C. difficile* infection was a Cochrane Review published in 2017 by Goldenberg et al. In this meta-analysis of 31 randomized controlled trials and greater than 8,500 patients, the incidence of *C. difficile* infection in those receiving prophylactic probiotics was 1.5% compared with 4.0% in the placebo or no treatment group. The probiotics were also shown to be safe in those who were not immune compromised or severely ill. So again, it seems that using *Saccharomyces boulardii* and other prophylactic probiotics has a significant benefit in preventing initial *C. difficile* episodes.

Alternatively, when considering usage of probiotics to prevent recurrence, this is a controversial topic with a couple of small studies showing favorable results and some others showing no benefit. In this space, there is a true shortage of well controlled trials and no absolute recommendations can be made based upon the literature. But in practice, I frequently consider using probiotics to prevent recurrence in those who are immune competent who do not have severe underlying illness.

Another novel approach to supplementing the microbiome to prevent *C. difficile* infection was presented by Dr. Dale Gerding several years ago. He had the truly novel idea of using a non-toxigenic form of *C. difficile*, meaning a form that is incapable of producing clinical illness, to support the microbiome as a way of minimizing recurrence after effective therapy. In this phase II placebo-controlled trial, 168 patients who were effectively treated for either
initial infection or first recurrence were included. They were assigned to three doses of oral non-toxigenic *C. difficile* or placebo. Interestingly, the group that received the non-toxigenic *C. difficile* had a recurrence rate of 11%, whereas those that received the placebo recurred 30% of the time.

Those are aggregate numbers, but when the non-toxigenic *C. difficile* infection was seen to formally engraft in the microbiome, meaning the patient took the non-toxigenic *C. difficile* and it stayed within their system, the recurrence rates actually plummeted to 2%. And, the engraftment rate was 69%. So, it’s believed that this non-toxigenic *C. difficile* can compete metabolically with the toxigenic strains, fill the niche, thereby potentially aiding the existing colonization resistance in the eradication of *C. difficile*. This of course is a very promising approach for supplementation of the microbiome to prevent recurrence of *C. difficile*, but we’re eagerly awaiting the phase III trials and larger studies associated with this.

**Question:** Thank you for that summary of the gut microbiome and the impact of available therapies. What about immune response? Are there ways to supplement the immune system response to our benefit?

**Dr. Feuerstadt:** As discussed with the Kyne studies previously, the immune response plays an important role in both minimizing initial onset of infection and decreasing risks for recurrence. Initially, intravenous immunoglobulin was used to try to minimize the risks for recurrence through a serologic immune stimulated effect. Results with this intravenous treatment were mixed, and one small prospective randomized controlled trial from about ten years ago showed no significant benefit from this. As a result, this has largely been abandoned.

However, in early 2017, a new, fully humanized monoclonal antibody called bezlotoxumab was approved by the FDA for usage in patients with *C. difficile*
infection at risk for recurrence. This infusion is given in addition to a standard of care antimicrobial and is not a solitary therapy. It is given as a one-time infusion that circulates within the bloodstream and then crosses the colonic mucosal barrier, accessing the lumen of the colon, binding to toxin B in a specific way and reducing the activity of the toxin. Its efficacy was seen in two large randomized controlled trials called MODIFY I and MODIFY II. These are placebo-controlled trials where in MODIFY I 386 individuals received bezlotoxumab and in MODIFY II 395 individuals received bezlotoxumab, in addition to an antimicrobial. Patients were then followed for three months.

A priori within these studies it was decided that there would be subgroup analyses based on risk factors for recurrence, including age over 65, a history of *Clostridium difficile* infection within the last six months, being in an immune compromised state, severe *C. difficile* infection based upon criteria from Zar et al, and whether the patient was infected with the hypervirulent NAP1/BI/027 strain. Overall, the group that received bezlotoxumab were 68% inpatients, 50% over 65, and 28% had *C. difficile* infection in the past six months. So, this was a real population.

Focusing on the primary endpoint of recurrence in MODIFY I, recurrence rates with placebo were 27.6%, compared with 17.4% in the bezlotoxumab group. In MODIFY II recurrence rates with placebo were 25.7% compared with 15.7% in the bezlotoxumab-treated group. Both were statistically significant differences with a number needed to treat of about ten. However, when the subgroup analyses were performed, there were trends for reduced recurrence rates in those that had a history of *C. difficile* infection, immune compromised, severe *C. difficile* infection, and the NAP1/BI/027 strain. There was a statistically significant reduction in both studies in recurrence in those over 65 years of age. The number needed to treat to reduce one recurrence in those over 65 was approximately six. So, we can see that in an appropriate population at
risk for recurrence, a product like bezlotoxumab given in addition to an antimicrobial can be an effective way to reduce future recurrence for these patients.

Question: This approach seems to meet an unmet clinical need. Are there any available data on the impact of bezlotoxumab on clinical outcomes?

Dr. Feuerstadt: Yes, there have been some recent analyses considering clinically relevant endpoints associated with bezlotoxumab in addition to recurrence. One study by Prabhu and colleagues performed a subgroup analysis from the original phase III randomized controlled trials MODIFY I and MODIFY II. When 781 bezlotoxumab-treated individuals were compared with 773 of those given placebo, 30-day readmission rates associated with *C. difficile* were significantly lower in the bezlotoxumab cohort compared with the placebo. Readmission rates were 5.1% with bezlotoxumab versus 11.2% with placebo. All-cause readmission rates were also lower, albeit non-statistically significantly, with the bezlotoxumab group having an all-cause readmission rate of 23.2% compared with 26.9% in placebo.

When considering the subgroups at highest risk for recurrence of *C. difficile* infection, those over 65 and those with severe infection were significantly less likely to be readmitted within 30 days for a recurrence of *C. difficile*, with trends across all of the other subgroups. Larger cohorts might further reinforce reduction in 30-day readmission rates with bezlotoxumab. The reduction of recurrence seen with this product along with reduced readmission rates can significantly improve patient outcomes and lessen the burden of this disease on both the patient themselves and, of course, the healthcare system.

It might be challenging to identify the ideal indication to use a treatment like bezlotoxumab. However, one study that provided a lot of insight into this
threshold was put forward by Dr. Gerding. He looked at the number of risk factors associated with recurrence and considered the rates of recurrence in those that received bezlotoxumab compared with placebo. When two risk factors were present, the rates of recurrence in those that received placebo were 41.1% compared with 26.9% in bezlotoxumab, a statistically significant difference. Whereas when three risk factors for recurrence were present, recurrence rates were 46.1% with placebo versus 21.2% with the bezlotoxumab. Given these statistically significant differences, as a clinician, one could approach patients with *C. difficile* by analyzing their risk factors for recurrence, and if the patients have two to three or more risk factors, there seems to be convincing evidence that the patient would benefit from this therapy.

Practically, though, once we decide the clinical scenarios where a patient might benefit from this treatment, it is also important to consider when to infuse bezlotoxumab. Within the original phase III randomized control trials, 94% of patients received the infusion within six days of initiation of the antimicrobial. In clinical practice, this product is, in theory, most active when the mucosa of the colon is most compromised, since the bezlotoxumab circulates in the bloodstream but crosses the mucosal barrier more readily when the mucosal barrier is compromised, and therefore it will have more access to bind toxin B. Practically speaking, there is a question of whether there might be a decrease in efficacy after longer durations of antimicrobials, since the mucosa theoretically is healing as the infection is progressively treated. Birch and colleagues considered this by comparing recurrence rates when bezlotoxumab was given within two days of a start of antimicrobials, between two and four days after or greater than five days following the initiation.

Within this study, there were no statistically significant differences when the three study groups were compared. Given this, as we approach patients that we think might benefit from bezlotoxumab, even if they have been on
antimicrobials for more than five days, so long as they are still on the antimicrobial, bezlotoxumab seems to perform very well in reducing the risks for recurrence. And this, follows the FDA indication for this product being infused during the antimicrobial course.

Question: Thank you for your time in this discussion. Can you please summarize your key takeaways for our learners?

Dr. Feuerstadt: Yes, so we have covered a broad set of topics during this question and answer. We’ve discussed two main mechanisms that the body uses to fight off \textit{C. difficile}: the microbiota and the serologic immune response. Understanding these two systems and these two responses will allow us as clinicians the ability to more efficiently treat \textit{C. difficile} infection. Specifically focusing on the microbiome, there is microbiota suppression that happens as a risk factor to initial infection, there’s collateral damage that happens with broader spectrum antimicrobials such as metronidazole and vancomycin, and then there’s the window of vulnerability, which is the timeframe from the completion of antimicrobial therapy to about three months into the future where colonization resistance is most active in wiping out the \textit{C. difficile}. Narrow spectrum antimicrobials, such as ridinilazole or fidaxomicin are able to treat \textit{C. difficile} while still preserving a lot of that colonization resistance, and therefore are associated with lower recurrence rates of the infection when used.

But there are also ways that we can supplement the microbiome and the colonization resistance, and that takes the form of probiotics, such as \textit{Saccharomyces boulardii}, that can be used to prophylactically minimize the risk for \textit{C. difficile} infection and its initial onset in individuals who are receiving antimicrobials for other indications. As we discussed before, the use of probiotics for recurrence is controversial, although I frequently will use these probiotics if the patients are immune competent and don’t have severe
underlying illness. Interestingly, one alternate probiotic might be using a non-toxigenic form of *C. difficile* to prevent *C. difficile* recurrence. And there are encouraging data showing that when we administer a non-toxigenic form, that we can actually prevent recurrence in a very, very strong way.

On the other side of things, we can also supplement the serologic or blood-borne immune system response, and that takes the form of the FDA-approved bezlotoxumab, a fully humanized monoclonal antibody to toxin B that is a onetime infusion given in addition to an antimicrobial in the appropriate populations. The data support using this product in patients that have two or three or more risk factors for recurrence. And the outcomes are very, very encouraging. Plus, the group over 65, who at high-risk for recurrence, can benefit from this.

So, we can see through this learning session that by understanding the microbiome and understanding the serologic response and understanding the options for treatment that we have for patients, we can be efficient in our therapy for *C. difficile* and optimize patients’ outcomes.