ANNUAL REVIEWS

Annual Review of Medicine

Microbiome Anomalies in Allogeneic Hematopoietic Cell Transplantation

Zaker I. Schwabkey[1] and Robert R. Jenq[1,2]

[1]Department of Genomic Medicine, Division of Cancer Medicine, The University of Texas MD Anderson Cancer Center, Houston, Texas 77030, USA

[2]Department of Stem Cell Transplantation and Cellular Therapy, Division of Cancer Medicine, The University of Texas MD Anderson Cancer Center, Houston, Texas 77030, USA; email: rrjenq@mdanderson.org

Annu. Rev. Med. 2020. 71:137–48

The *Annual Review of Medicine* is online at med.annualreviews.org

https://doi.org/10.1146/annurev-med-052918-122440

Copyright © 2020 by Annual Reviews. All rights reserved

ANNUAL REVIEWS CONNECT

www.annualreviews.org

- Download figures
- Navigate cited references
- Keyword search
- Explore related articles
- Share via email or social media

Keywords

allogeneic hematopoietic cell transplant, allo HCT, graft-versus-host disease, GVHD, microbiome, antibiotics, transplant-related mortality, TRM, febrile neutropenia

Abstract

The microbiome is an integrated part of the human body that can modulate a variety of disease processes and affect prognosis, treatment response, complications, and outcomes. The importance of allogeneic hematopoietic cell transplantation in cancer treatment has resulted in extensive investigations on the interaction between the microbiome and this treatment modality. These investigations are beginning to lead to clinical trials of microbiome-targeted interventions. Here we review some of these discoveries and describe strategies being investigated to manipulate the microbiome for favorable outcomes, such as the proper selection and timing of antibiotics, type of diet and route of administration, probiotics, prebiotics, and fecal microbiota transplantation.

INTRODUCTION

Accumulating evidence demonstrates that the community of commensal bacteria residing in our gastrointestinal (GI) tract is an important modulator of disease at biochemical and molecular levels (1). The Human Microbiome Project (2) uncovered a surprising degree of microbiome variability between healthy individuals and has been a great tool, paving the way to help researchers investigate how disease parameters and clinical outcomes are related to microbiome heterogeneity, both at baseline and as disease advances.

The evidence of an association has been particularly convincing for intestinal inflammatory diseases, including ulcerative colitis, Crohn's disease, and graft-versus-host disease (GVHD), which is an immune-mediated condition mediated by cells derived from the graft infused into the recipient of an allogeneic hematopoietic cell transplant (allo HCT) (3). Given that these are all intestinal inflammatory syndromes, the question arises of whether close proximity is an important contributor to microbiome–disease interactions.

MICROBIOTA DAMAGE IS COMMON IN ALLOGENEIC HEMATOPOIETIC CELL TRANSPLANTATION

Allo HCT is a lengthy treatment modality that often results in multiple insults to the recipient's microbiome structure. One way this structure can be characterized is by quantifying alpha diversity, an ecological measure that combines levels of richness of distinct bacteria and evenness in their relative abundances. Alpha diversity is commonly used as a convenient measure of microbiome health and is frequently reduced in patients with disease states. Longitudinal monitoring of the microbiome of allo HCT recipients has demonstrated that changes in the microbiome, including loss of diversity, can be observed early during the treatment course when patients receive chemotherapeutic agents, radiation, and the donor's hematopoietic cells (4). Reduced diversity is often accompanied by prominent decreases in the abundances of commensal intestinal bacteria, many of which have been shown to have beneficial contributions to normal intestinal homeostasis including digestion, barrier function, and immune tolerance (5).

MICROBIOTA CHANGES ARE ASSOCIATED WITH VARIOUS POOR OUTCOMES

The intestinal microbiome during and after allo HCT has been examined by several groups with largely concordant results. Many adverse outcomes are strongly associated with dysbiosis of the gut microbiota. Here we summarize some of these outcomes and how the microbiota is affected.

Graft-Versus-Host Disease

Intestinal GVHD is common after allo HCT, and inflammation is the hallmark of this disease. Loss of bacterial diversity was noted to occur early after allo HCT in mice with GVHD (6). Different mechanisms may explain microbiota contributions to GVHD. α-Defensins are antimicrobial peptides produced by specialized granule-harboring epithelial cells known as Paneth cells that can target potentially pathogenic bacteria. In GVHD, Paneth cells are injured, resulting in lower expression of antimicrobial peptides and possibly predisposing to dysbiosis by allowing harmful bacteria to expand (7).

Febrile neutropenia in patients undergoing allo HCT is common and requires early treatment with empiric broad-spectrum antibiotics. Several of these antibiotics are considered clinically equivalent but can vary in their degree of collateral damage to the microbiome (discussed in

detail later in this review). Shono et al. (8) found that GVHD severity, along with microbiome damage, can differ depending on the type of antibiotic used in both humans and mice. Specifically, antibiotics with stronger activity against obligate anaerobes, including piperacillin-tazobactam and carbapenems, resulted in increased disruption to the microbiome, higher intestinal barrier damage, and more severe GVHD, compared to a narrower-spectrum antibiotic such as aztreonam.

Congruent with these antibiotic associations, multiple studies have found that preservation of obligately anaerobic bacteria is associated with protection from GVHD. Abundance of bacteria from the genus *Blautia*, an abundant commensal member of the Clostridia class of Firmicutes, has been associated with better survival, less GVHD-related or relapse-related mortality, and less need for GVHD treatment with systemic steroids (9). An independent study of adult patients also found that bacteria from the family Lachnospiraceae (which includes *Blautia* and belongs to the class Clostridia) are associated with less severe GVHD (10). Pediatric patients with Clostridia were also found to have reduced GVHD in independent cohorts from two transplant centers (11). Mechanisms for this association remain unclear, but one study found that recovery of mucosal-associated invariant T cells, an immune regulatory population, is highly heterogeneous after allo HCT but is associated with increased abundance of intestinal *Blautia* species (12). A metabolite commonly produced by Clostridia, butyrate, was found to directly alleviate GVHD in mouse models, via support of intestinal epithelial cells (13). In addition to Clostridia, other clinical studies have found that *Bacteroides*, another intestinal obligately anaerobic commensal, was less abundant in GVHD (14). One open question is whether, in addition to the host microbiome, the composition of the HCT donor's microbiome can have any potential impact on GVHD severity.

Clostridium difficile Infection

Clostridium difficile infection (CDI) in allo HCT patients requires close monitoring. Watery diarrhea is often the presenting symptom that prompts testing, but sometimes treatment can be started before test results are known (15). A single-center retrospective study showed that the majority of CDI cases occur in the first six months following allo HCT. Total-body irradiation and GVHD are known risk factors as both result in gut dysbiosis and intestinal barrier dysfunction, predisposing to local infection and bacteremia, respectively, with less clear effects on overall mortality (16).

The majority of cases of CDI occur in the peritransplantation period. Healthier patients without comorbid diseases are less likely to develop CDI in the pre-engraftment period, but CDI in the postengraftment period is common and may potentially warrant outpatient surveillance (17), though this is not yet recommended by guidelines. Presence at engraftment of usually abundant obligate anaerobes, including Bacteroidetes, Lachnospiraceae, and Ruminococcaceae, was associated with a 60% lower risk of CDI in the postengraftment period (18). Since antibiotics are routinely administered during the course of allo HCT, CDI in this patient population frequently recurs and is associated with loss of the same obligately anaerobic intestinal bacteria (19).

Bacteremia

Bacteremia is a potentially lethal complication in patients undergoing allo HCT. In patients who develop this complication, reduced microbiome diversity is frequently observed (20). They also display loss of obligately anaerobic commensal bacteria and instead often show gut domination by pathogenic bacteria, including *Enterococcus* and Proteobacteria (the phylum that includes *Escherichia coli*, *Klebsiella*, and other gram-negative facultatively anaerobic bacteria), which match the

bacteria isolated from the bloodstream. This observation suggests that the pathophysiology of bacteremia may require dysbiosis as well as impaired intestinal barrier function. Domination with *Enterococcus*, including strains expressing genes that encode vancomycin resistance, has been particularly associated with metronidazole use. Interestingly, Proteobacteria domination was reduced by levofloxacin use, an example where antibiotics can have a positive clinical effect via the microbiome. In another study, a broadly active combination of ciprofloxacin and metronidazole prophylaxis was associated with domination by *Enterococcus*, and the domination was more pronounced when GVHD was present (21). This emphasizes the importance of antibiotic selection in the peritransplantation period, further discussed below.

Recently, high-resolution metagenomic sequencing of paired blood and stool samples from HCT patients with bacteremia confirmed that in many but not all cases, commensal intestinal bacteria and blood pathogens shared significant genetic similarity, indicating a highly probable gut origin (22). Interestingly, this was also true for some pathogens that are less certain to be enteric, such as *Pseudomonas aeruginosa* and *Staphylococcus epidermidis*, suggesting that the traditional teaching that these pathogens are normally rare in the intestinal tract is not necessarily true in all patient populations.

Respiratory Complications

The upper respiratory tract hosts the majority of the microbiome that can potentially relocate to the lungs, and associations between the intestinal microbiome and pulmonary outcomes after allo HCT have also been observed. Challenges in studying the microbiome of the respiratory tract include variability in sampling methods (23), but significant progress is being made. Parallel RNA/DNA sequencing of lower respiratory specimens in immune-compromised pediatric patients could identify pathogens in many patients where no clinical isolate could be identified using current microbiological methods, indicating that sequencing approaches can augment standard techniques (24). Interestingly, evidence for a gut–lung axis also exists. A study by Harris et al. (25) showed that low baseline GI microbiome diversity at the time of engraftment was associated with more early pulmonary complications including infections, transfusion reactions, and interstitial pneumonitis. Peritransplant antibiotic use has been found to be a risk factor for development of respiratory viral infections in allo HCT recipients (26), and GI colonization with butyrate-producing commensal bacteria was associated with protection from lower respiratory tract viral infection following allo HCT (27), suggesting that intestinal bacteria–derived butyrate could potentially support systemic antiviral immunity. Similarly, gut domination of Proteobacteria in allo HCT recipients was associated with low survival and more respiratory complications (28).

Overall Survival

Studies have shown that intestinal microbiota diversity at the time of engraftment is a potential clinical predictor of mortality in allo HCT (29). This association appears to be driven by treatment-related mortality (TRM), a composite endpoint that includes GVHD-related mortality and mortality due to infectious complications, which have individually been associated with microbiome damage as described above and are often inextricably linked clinically. Cytomegalovirus, for example, reactivates frequently in patients treated with immune suppression for GVHD, but reactivation can also precede GVHD (30). Notably, TRM excludes relapse, and relapse as an adverse outcome has not been associated with reduced microbiome diversity (31). Potential relationships that exist between disease relapse following allo HCT and host microbiome are understudied. Peled et al. (31) have reported an association between presence or higher abundance of

Eubacterium limosum and decreased risk for relapse in these patients. Interestingly, not all disease types were equally associated; freedom from relapse of acute myelogenous leukemia and multiple myeloma were particularly associated with having *Eubacterium limosum* in the intestinal tract. Whether this observation can be replicated by other centers is still unknown.

THE MICROBIOME AS A TARGET OF INTERVENTION

The observation that the intestinal microbiome composition is associated with a variety of important clinical outcomes after allo HCT, including that the healthy microbiome diversity often precedes any TRM from allo HCT, leads to hypotheses that have yet to be formally tested clinically. One is that loss of diversity is a biomarker that can robustly and reproducibly predict survival after allo HCT. Multicenter efforts are currently under way to address this hypothesis, including a trial by the Blood and Marrow Transplant Clinical Trials Network (BMT CTN 1703/1801) (NCT03959241). A second, potentially more intriguing hypothesis is that damage to the intestinal microbiome can be prevented or treated, and this may lead to improved clinical outcomes.

Reducing the complexity of microbiome data to an alpha diversity metric is very useful, given the relative ease of intuitive understanding, as well as the strength of the metric's association with outcomes such as overall survival. There are important limitations, however, to using alpha diversity as a biomarker (32). Some are methodological—there are many ways to quantify and weigh the relative contributions of evenness and richness, and it is unclear which methods are more predictive of outcomes. There are also upstream computational decisions that would have to be standardized to allow characterization of a normal range of diversity, including when and how to rarefy sequences to deal with inherent differences in sequencing depth. It is also possible for the metric to yield "false positives"—for example, if many normal commensal bacteria are missing, but bacteria that are present happen to be equally abundant, diversity will technically be high despite obvious dysbiosis. Finally, current sequencing technologies require batching hundreds of samples in order for 16S sequencing to be cost-effective, and thus a quick turnaround time that would be necessary for microbiome diversity to guide clinical practice is not yet feasible.

An alternative to directly measuring microbiome diversity by sequencing is to detect its metabolic activity. Weber et al. (33) developed a tool that can indirectly quantify microbiota composition by measuring levels of 3-indoxyl sulfate in the urine. This metabolite is produced from indole that is a by-product of metabolism of dietary tryptophan by intestinal bacteria. The authors found that higher levels of urinary 3-indoxyl sulfate early after transplantation predicted reduced transplant-related mortality and improved overall survival.

Optimizing Antibiotic Use

Antibiotics are used frequently in allo HCT patients, whether prescribed empirically for febrile neutropenia or culture-targeted for known infections. The practice of using antibiotics in general focuses on eradicating or preventing pathogenic bacteria with minimal attention on the commensal bacteria.

Empiric and targeted treatment for neutropenic fever. Current guidelines for the treatment of neutropenic fever do not take into consideration microbiome bystander effects, and many recommended antibiotics are in fact highly damaging to obligately anaerobic intestinal commensals. Shono et al. (8) compared common antibiotics used for febrile neutropenia and found that imipenem-cilastatin and piperacillin-tazobactam were associated with higher GVHD-related mortality than aztreonam and cefepime. Mice experiments showed that imipenem-treated mice

had aggravated GVHD, and this was attributable to the expansion of intestinal mucus-degrading bacteria *Akkermansia muciniphila*. Holler et al. (21) found that gut dysbiosis characterized by *Enterococcus* predominance was commonly seen in patients receiving ciprofloxacin and metronidazole prophylaxis. In pediatric patients undergoing allo HCT, exposure to clindamycin was associated with depletion of Clostridia and an increased likelihood of GVHD (11). A recent retrospective review found that patients who received oral broad-spectrum antibiotics as bacterial prophylaxis had higher rates of GVHD (34). Finally, the timing of antibiotic administration has also been retrospectively associated with GVHD; patients who began receiving antibiotics for neutropenic fever pretransplant had worse outcomes than those who received antibiotics post-transplant, with reductions in stool Clostridia abundance and urinary 3-indoxyl sulfate as well as increased TRM (35).

A randomized clinical trial is currently investigating how the use of piperacillin-tazobactam and cefepime as empiric antibiotic therapy for febrile neutropenia in allo HCT can affect Clostridiales abundance in the intestine (NCT03078010).

Prophylactic strategies: oral decontamination. In contrast to recent studies indicating that antibiotic use leads to microbiome injury and aggravates GVHD, early studies in the 1970s in mice found that gut decontamination with oral antibiotics reduced GVHD and improved survival (36). Initial clinical studies showed similar benefits (37), as did a more recent randomized study (38). Results of subsequent studies, however, have been mixed (39). A recent retrospective microbiological analysis of stool specimens collected from patients who received decontamination found that the success rate of decontamination was only roughly 50%, and successful decontamination was associated with less acute GVHD (40). A clinical trial is planned to re-evaluate oral decontamination followed by third-party fecal microbiota transplantation (FMT) in selected allo HCT patients who require antibiotic treatment with meropenem or piperacillin-tazobactam, both of which have been associated with increased GVHD (NCT03862079).

Prophylactic strategies: selective decontamination. Per joint American Society of Clinical Oncology/Infectious Diseases Society of America guidelines, fluoroquinolones are the preferred prophylaxis in febrile neutropenia (41). Levofloxacin was very effective in reducing infections and bacteremia in patients with high-risk neutropenia (42). More recent studies have found that quinolone prophylaxis is associated with reduced intestinal domination by gram-negative bacteria (20). Thus, quinolones appear to mediate a benefit by selectively reducing potentially proinflammatory bacteria without broadly targeting obligately anaerobic commensals. Polymixin B, which similarly targets gram-negative bacteria, showed a similar benefit in a mouse model of GVHD by reducing the abundance of intestinal *E. coli* (7).

A possible alternative to fluoroquinolones for bacterial prophylaxis allo HCT is rifaximin. The transplant center at Regensburg changed its institutional practice from prophylaxis with ciprofloxacin and metronidazole to rifaximin, and compared outcomes in two cohorts of patients before and after the switch (43). Investigators found comparable episodes of fever, infections, and bacteremia, suggesting similar efficacy for infectious prophylaxis. Intriguingly, they also noted that patients receiving rifaximin had reduced gut dysbiosis with higher urinary 3-indoxyl sulfate, as well as reduced transplant-related mortality and improved overall survival.

Microbiome Interventions

In addition to optimizing antibiotics use, other strategies have been studied to manipulate the microbiome. Some of them have been proven to be safe with favorable TRM outcomes, such as selection of diet, probiotics, and prebiotics, and others have reported complications despite promising outcomes, such as FMT.

Diet. When medically not contraindicated, oral intake is usually encouraged in allo HCT patients, and clinicians monitor oral nutrition as a measure of a healthy prognosis, including a reduced incidence of severe GVHD (44). One study showed that in allo HCT patients, supplemental enteral nutrition, compared to parenteral nutrition, was associated with better overall survival and protection from severe acute GVHD (45). Similar results were found with home-based care for allo HCT as these patients had better oral intake than those transplanted in the inpatient hospital setting (46). Another study showed that parenteral nutrition in allo HCT patients, compared to enteral feeding, was associated with more infections, increased escalation of medical care with central access, and transfer to intensive care, as well as higher overall mortality from infections (47, 48). One limitation on drawing conclusions from nonrandomized studies associating improved oral nutrition with better outcomes is that patients who are able to tolerate oral intake are generally healthier and more willing to initiate oral feeding than parenterally fed patients. A phase III clinical trial by a group in France is currently comparing enteral to parenteral nutrition (NCT01955772).

The type of oral nutrition in the peri-HCT period may also be an important factor. Traditionally, allo HCT patients are recommended to limit their diet to well-cooked foods and peeled fruits, known as a neutropenic diet. In recent years, this practice has been examined and been found to have a limited benefit, or even potentially some harm (49). One center recently made an institutional practice switch from a neutropenic diet to a more liberalized diet and found no increase in infections or other adverse outcomes (50). A randomized study addressing the same question also found no benefit to a neutropenic diet (51). The short-term (52) and long-term (53) effects of dietary differences on the microbiome have been reported in healthy volunteers, and the effects in the allo HCT population warrant further study.

Prebiotics. Prebiotics are dietary supplements designed to promote the growth or modify the metabolism of beneficial intestinal commensal bacteria, and are commonly used to target the microbiome. In allo HCT patients, a small number of studies have been performed. One study in Japan showed that allo HCT patients who were given supplemental products enriched with glutamine, fiber, and oligosaccharides (polydextrose, lactosucrose, and dextrin) had less diarrhea, mucositis, and *Enterococcus* bacteremia, with better survival (54). Potato starch (NCT02763033), a gluten-free diet (NCT03102060), and fructose oligosaccharides (NCT02805075) are each currently being studied for potential benefit in HCT patients.

Probiotics. Probiotics are ingestible formulations of live bacteria that can modulate intestinal homeostasis. Because they are regulated only for safety and not for medical efficacy, they are widely available, and many questions regarding their effects on the microbiome and health persist (55). Some probiotic studies have been performed in the allo HCT population. *Lactobacillus plantarum* was recently found to be safe to administer during neutropenia in the pediatric allo HCT population (56), and a multicenter study evaluating this probiotic for potential benefit for GVHD is under way (NCT03057054).

Results with *Lactobacillus rhamnosus* GG (LGG) have been mixed. In a murine model, administration of LGG resulted in lower GVHD scores, milder pathologic sequelae, and improved survival (57). A recent randomized clinical trial, however, did not show any appreciable protection from GVHD in humans receiving LGG (58). Patients receiving LGG did not show measurable increases in the abundance of *Lactobacillus*, a finding that is in accordance with prior studies in healthy volunteers indicating lack of persistent colonization, though colonoscopic biopsies appear to be more sensitive for detecting LGG than stool samples (59).

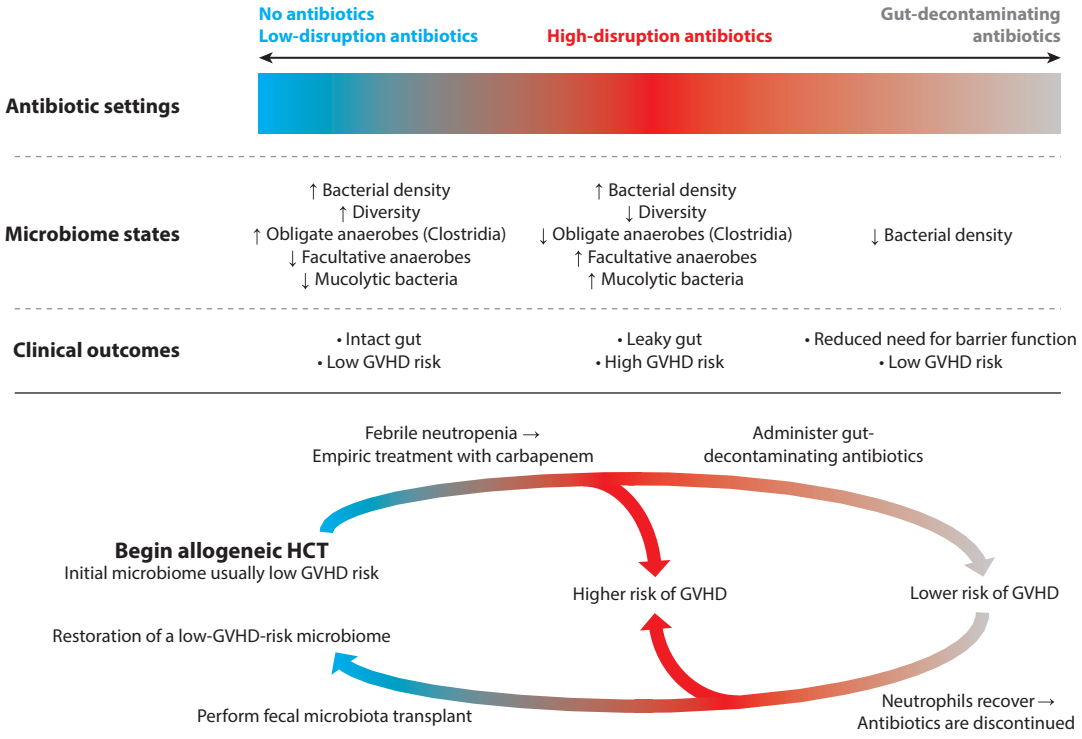

Figure 1

Empiric antibiotics used for febrile neutropenia in hematopoietic cell transplant (HCT) patients have been linked to a dysbiotic microbiome and worsened outcomes, including increased graft-versus-host disease (GVHD). Fecal microbiota transplantation can potentially correct this disruption and potentially reduce GVHD.

Fecal microbiota transplantation. FMT has been reliably shown to be effective in treating recurrent CDI (60, 61), including in allo HCT patients with recurrent CDI (62–64). In the allo HCT population, FMT has also been performed as therapy for GVHD, reported now in several small case series (65–68). In all of these, patients with steroid-dependent or steroid-refractory GVHD responded to FMT and their microbiome composition improved. The treatment was generally well tolerated without serious adverse effects.

The prophylactic use of FMT in allo HCT recipients without symptoms of GVHD has also begun to be investigated by two groups, including a 13-patient single-arm study of oral frozen third-party FMT capsules (69) and a randomized study of 25 patients, 14 of whom received an autologous FMT collected prior to HCT conditioning (70). Both studies administered FMT following neutrophil recovery, and both show evidence of improved microbiome composition in patients following FMT (**Figure 1**). However, there is not yet any evidence of improvements in clinical outcomes with prophylactic FMT, and follow-up as well as larger studies are being planned.

CONCLUSION

Early insights tend to revive as development of new technologies allows a better understanding of patterns observed in the past. The recent discovery of changes in the microbiome during allo HCT is a perfect example of this phenomenon. The importance of clinical decision making that

incorporates cognizance of the microbiome is increasingly apparent, and additional strategies to protect, restore, and perhaps even augment the microbiome may yet provide approaches to further optimize outcomes in this vulnerable patient population.

DISCLOSURE STATEMENT

Dr. Jenq serves on the advisory boards of Seres Therapeutics, Inc., and Kaleido Biosciences, Inc., and holds intellectual property rights in Seres Therapeutics, Inc. He serves as a consultant to Ziopharm Oncology, MicrobiomeDx, Merck, and Novartis. Dr. Schwabkey is not aware of any affiliations, memberships, funding, or financial holdings that might be perceived as affecting the objectivity of this review.

LITERATURE CITED

1. Slingerland AE, Schwabkey Z, Wiesnoski DH, Jenq RR. 2017. Clinical evidence for the microbiome in inflammatory diseases. *Front. Immunol.* 8:400
2. Human Microbiome Project Consortium. 2012. Structure, function and diversity of the healthy human microbiome. *Nature* 486(7402):207–14
3. Eckburg PB, Bik EM, Bernstein CN, et al. 2005. Diversity of the human intestinal microbial flora. *Science* 308(5728):1635–38
4. Taur Y. 2016. Intestinal microbiome changes and stem cell transplantation: lessons learned. *Virulence* 7(8):930–38
5. Montassier E, Batard E, Massart S, et al. 2014. 16S rRNA gene pyrosequencing reveals shift in patient faecal microbiota during high-dose chemotherapy as conditioning regimen for bone marrow transplantation. *Microb. Ecol.* 67(3):690–99
6. Jenq RR, Ubeda C, Taur Y, et al. 2012. Regulation of intestinal inflammation by microbiota following allogeneic bone marrow transplantation. *J. Exp. Med.* 209(5):903–11
7. Eriguchi Y, Takashima S, Oka H, et al. 2012. Graft-versus-host disease disrupts intestinal microbial ecology by inhibiting Paneth cell production of alpha-defensins. *Blood* 120(1):223–31
8. Shono Y, Docampo MD, Peled JU, et al. 2016. Increased GVHD-related mortality with broad-spectrum antibiotic use after allogeneic hematopoietic stem cell transplantation in human patients and mice. *Sci. Transl. Med.* 8(339):339ra371
9. Jenq RR, Taur Y, Devlin SM, et al. 2015. Intestinal *Blautia* is associated with reduced death from graft-versus-host disease. *Biol. Blood Marrow Transplant.* 21(8):1373–83
10. Golob JL, Pergam SA, Srinivasan S, et al. 2017. Stool microbiota at neutrophil recovery is predictive for severe acute graft versus host disease after hematopoietic cell transplantation. *Clin. Infect. Dis.* 65(12):1984–91
11. Simms-Waldrip TR, Sunkersett G, Coughlin LA, et al. 2017. Antibiotic-induced depletion of anti-inflammatory Clostridia is associated with the development of graft-versus-host disease in pediatric stem cell transplantation patients. *Biol. Blood Marrow Transplant.* 23(5):820–29
12. Bhattacharyya A, Hanafi LA, Sheih A, et al. 2018. Graft-derived reconstitution of mucosal-associated invariant T cells after allogeneic hematopoietic cell transplantation. *Biol. Blood Marrow Transplant.* 24(2):242–51
13. Mathewson ND, Jenq R, Mathew AV, et al. 2016. Gut microbiome-derived metabolites modulate intestinal epithelial cell damage and mitigate graft-versus-host disease. *Nat. Immunol.* 17(5):505–13
14. Biagi E, Zama D, Nastasi C, et al. 2015. Gut microbiota trajectory in pediatric patients undergoing hematopoietic SCT. *Bone Marrow Transplant.* 50(7):992–98
15. McDonald LC, Gerding DN, Johnson S, et al. 2018. Clinical practice guidelines for *Clostridium difficile* infection in adults and children: 2017 update by the Infectious Diseases Society of America (IDSA) and Society for Healthcare Epidemiology of America (SHEA). *Clin. Infect. Dis.* 66(7):e1–48

16. Willems L, Porcher R, Lafaurie M, et al. 2012. *Clostridium difficile* infection after allogeneic hematopoietic stem cell transplantation: incidence, risk factors, and outcome. *Biol. Blood Marrow Transplant.* 18(8):1295–301
17. Dubberke ER, Reske KA, Olsen MA, et al. 2017. Risk for *Clostridium difficile* infection after allogeneic hematopoietic cell transplant remains elevated in the postengraftment period. *Transplant. Direct.* 3(4):e145
18. Lee YJ, Arguello ES, Jenq RR, et al. 2017. Protective factors in the intestinal microbiome against *Clostridium difficile* infection in recipients of allogeneic hematopoietic stem cell transplantation. *J. Infect. Dis.* 215(7):1117–23
19. Chang JY, Antonopoulos DA, Kalra A, et al. 2008. Decreased diversity of the fecal microbiome in recurrent *Clostridium difficile*-associated diarrhea. *J. Infect. Dis.* 197(3):435–38
20. Taur Y, Xavier JB, Lipuma L, et al. 2012. Intestinal domination and the risk of bacteremia in patients undergoing allogeneic hematopoietic stem cell transplantation. *Clin. Infect. Dis.* 55(7):905–14
21. Holler E, Butzhammer P, Schmid K, et al. 2014. Metagenomic analysis of the stool microbiome in patients receiving allogeneic stem cell transplantation: loss of diversity is associated with use of systemic antibiotics and more pronounced in gastrointestinal graft-versus-host disease. *Biol. Blood Marrow Transplant.* 20(5):640–45
22. Tamburini FB, Andermann TM, Tkachenko E, et al. 2018. Precision identification of diverse bloodstream pathogens in the gut microbiome. *Nat. Med.* 24(12):1809–14
23. Man WH, de Steenhuijsen Piters WA, Bogaert D. 2017. The microbiota of the respiratory tract: gatekeeper to respiratory health. *Nat. Rev. Microbiol.* 15(5):259–70
24. Zinter MS, Dvorak CC, Mayday MY, et al. 2019. Pulmonary metagenomic sequencing suggests missed infections in immunocompromised children. *Clin. Infect. Dis.* 68(11):1847–55
25. Harris B, Morjaria SM, Littmann ER, et al. 2016. Gut microbiota predict pulmonary infiltrates after allogeneic hematopoietic cell transplantation. *Am. J. Respir. Crit. Care Med.* 194(4):450–63
26. Ogimi C, Krantz EM, Golob JL, et al. 2018. Antibiotic exposure prior to respiratory viral infection is associated with progression to lower respiratory tract disease in allogeneic hematopoietic cell transplant recipients. *Biol. Blood Marrow Transplant.* 24(11):2293–301
27. Haak BW, Littmann ER, Chaubard JL, et al. 2018. Impact of gut colonization with butyrate-producing microbiota on respiratory viral infection following allo-HCT. *Blood* 131(26):2978–86
28. Bunker JJ, Flynn TM, Koval JC, et al. 2015. Innate and adaptive humoral responses coat distinct commensal bacteria with immunoglobulin A. *Immunity* 43(3):541–53
29. Taur Y, Jenq RR, Perales MA, et al. 2014. The effects of intestinal tract bacterial diversity on mortality following allogeneic hematopoietic stem cell transplantation. *Blood* 124(7):1174–82
30. Cantoni N, Hirsch HH, Khanna N, et al. 2010. Evidence for a bidirectional relationship between cytomegalovirus replication and acute graft-versus-host disease. *Biol. Blood Marrow Transplant.* 16(9):1309–14
31. Peled JU, Devlin SM, Staffas A, et al. 2017. Intestinal microbiota and relapse after hematopoietic-cell transplantation. *J. Clin. Oncol.* 35(15):1650–59
32. Wagner BD, Grunwald GK, Zerbe GO, et al. 2018. On the use of diversity measures in longitudinal sequencing studies of microbial communities. *Front. Microbiol.* 9:1037
33. Weber D, Oefner PJ, Hiergeist A, et al. 2015. Low urinary indoxyl sulfate levels early after transplantation reflect a disrupted microbiome and are associated with poor outcome. *Blood* 126(14):1723–28
34. Routy B, Letendre C, Enot D, et al. 2017. The influence of gut-decontamination prophylactic antibiotics on acute graft-versus-host disease and survival following allogeneic hematopoietic stem cell transplantation. *Oncoimmunology* 6(1):e1258506
35. Weber D, Jenq RR, Peled JU, et al. 2017. Microbiota disruption induced by early use of broad-spectrum antibiotics is an independent risk factor of outcome after allogeneic stem cell transplantation. *Biol. Blood Marrow Transplant.* 23(5):845–52
36. Jones JM, Wilson R, Bealmear PM. 1971. Mortality and gross pathology of secondary disease in germfree mouse radiation chimeras. *Radiat. Res.* 45(3):577–88

37. Storb R, Prentice RL, Buckner CD, et al. 1983. Graft-versus-host disease and survival in patients with aplastic anemia treated by marrow grafts from HLA-identical siblings. Beneficial effect of a protective environment. *N. Engl. J. Med.* 308(6):302–7
38. Beelen DW, Elmaagacli A, Muller KD, et al. 1999. Influence of intestinal bacterial decontamination using metronidazole and ciprofloxacin or ciprofloxacin alone on the development of acute graft-versus-host disease after marrow transplantation in patients with hematologic malignancies: final results and long-term follow-up of an open-label prospective randomized trial. *Blood* 93(10):3267–75
39. Petersen FB, Buckner CD, Clift RA, et al. 1986. Laminar air flow isolation and decontamination: a prospective randomized study of the effects of prophylactic systemic antibiotics in bone marrow transplant patients. *Infection* 14(3):115–21
40. Vossen JM, Guiot HF, Lankester AC, et al. 2014. Complete suppression of the gut microbiome prevents acute graft-versus-host disease following allogeneic bone marrow transplantation. *PLOS ONE* 9(9):e105706
41. Taplitz RA, Kennedy EB, Bow EJ, et al. 2018. Antimicrobial prophylaxis for adult patients with cancer-related immunosuppression: ASCO and IDSA clinical practice guideline update. *J. Clin. Oncol.* 2018:JCO1800374
42. Bucaneve G, Micozzi A, Menichetti F, et al. 2005. Levofloxacin to prevent bacterial infection in patients with cancer and neutropenia. *N. Engl. J. Med.* 353(10):977–87
43. Weber D, Oefner PJ, Dettmer K, et al. 2016. Rifaximin preserves intestinal microbiota balance in patients undergoing allogeneic stem cell transplantation. *Bone Marrow Transplant*. 51(8):1087–92
44. Bechard LJ, Guinan EC, Feldman HA, et al. 2007. Prognostic factors in the resumption of oral dietary intake after allogeneic hematopoietic stem cell transplantation (HSCT) in children. *J. Parenter. Enteral Nutr.* 31(4):295–301
45. Seguy D, Duhamel A, Rejeb MB, et al. 2012. Better outcome of patients undergoing enteral tube feeding after myeloablative conditioning for allogeneic stem cell transplantation. *Transplantation* 94(3):287–94
46. Svahn BM, Remberger M, Heijbel M, et al. 2008. Case-control comparison of at-home and hospital care for allogeneic hematopoietic stem-cell transplantation: the role of oral nutrition. *Transplantation* 85(7):1000–7
47. Seguy D, Berthon C, Micol JB, et al. 2006. Enteral feeding and early outcomes of patients undergoing allogeneic stem cell transplantation following myeloablative conditioning. *Transplantation* 82(6):835–39
48. Guieze R, Lemal R, Cabrespine A, et al. 2014. Enteral versus parenteral nutritional support in allogeneic haematopoietic stem-cell transplantation. *Clin. Nutr.* 33(3):533–38
49. Trifilio S, Helenowski I, Giel M, et al. 2012. Questioning the role of a neutropenic diet following hematopoetic stem cell transplantation. *Biol. Blood Marrow Transplant.* 18(9):1385–90
50. Taggart C, Neumann N, Alonso PB, et al. 2019. Comparing a neutropenic diet to a food safety-based diet in pediatric patients undergoing hematopoietic stem cell transplantation. *Biol. Blood Marrow Transplant.* 18(9):1385–90
51. Moody KM, Baker RA, Santizo RO, et al. 2018. A randomized trial of the effectiveness of the neutropenic diet versus food safety guidelines on infection rate in pediatric oncology patients. *Pediatr. Blood Cancer* 65(1):e26711
52. David LA, Maurice CF, Carmody RN, et al. 2014. Diet rapidly and reproducibly alters the human gut microbiome. *Nature* 505(7484):559–63
53. Wu GD, Chen J, Hoffmann C, et al. 2011. Linking long-term dietary patterns with gut microbial enterotypes. *Science* 334(6052):105–8
54. Iyama S, Sato T, Tatsumi H, et al. 2014. Efficacy of enteral supplementation enriched with glutamine, fiber, and oligosaccharide on mucosal injury following hematopoietic stem cell transplantation. *Case Rep. Oncol.* 7(3):692–99
55. Suez J, Zmora N, Segal E, Elinav E. 2019. The pros, cons, and many unknowns of probiotics. *Nat. Med.* 25(5):716–29
56. Ladas EJ, Bhatia M, Chen L, et al. 2016. The safety and feasibility of probiotics in children and adolescents undergoing hematopoietic cell transplantation. *Bone Marrow Transplant*. 51(2):262–66

57. Gerbitz A, Schultz M, Wilke A, et al. 2004. Probiotic effects on experimental graft-versus-host disease: let them eat yogurt. *Blood* 103(11):4365–67
58. Gorshein E, Wei C, Ambrosy S, et al. 2017. *Lactobacillus rhamnosus* GG probiotic enteric regimen does not appreciably alter the gut microbiome or provide protection against GVHD after allogeneic hematopoietic stem cell transplantation. *Clin. Transplant.* 31(5):e12947
59. Alander M, Satokari R, Korpela R, et al. 1999. Persistence of colonization of human colonic mucosa by a probiotic strain, *Lactobacillus rhamnosus* GG, after oral consumption. *Appl. Environ. Microbiol.* 65(1):351–54
60. Kassam Z, Lee CH, Yuan Y, Hunt RH. 2013. Fecal microbiota transplantation for *Clostridium difficile* infection: systematic review and meta-analysis. *Am. J. Gastroenterol.* 108(4):500–8
61. van Nood E, Vrieze A, Nieuwdorp M, et al. 2013. Duodenal infusion of donor feces for recurrent *Clostridium difficile*. *N. Engl. J. Med.* 368(5):407–15
62. Moss EL, Falconer SB, Tkachenko E, et al. 2017. Long-term taxonomic and functional divergence from donor bacterial strains following fecal microbiota transplantation in immunocompromised patients. *PLOS ONE* 12(8):e0182585
63. Bluestone H, Kronman MP, Suskind DL. 2018. Fecal microbiota transplantation for recurrent *Clostridium difficile* infections in pediatric hematopoietic stem cell transplant recipients. *J. Pediatr. Infect. Dis. Soc.* 7(1):e6–e8
64. Webb BJ, Brunner A, Ford CD, et al. 2016. Fecal microbiota transplantation for recurrent *Clostridium difficile* infection in hematopoietic stem cell transplant recipients. *Transpl. Infect. Dis.* 18(4):628–33
65. Kakihana K, Fujioka Y, Suda W, et al. 2016. Fecal microbiota transplantation for patients with steroid-resistant acute graft-versus-host disease of the gut. *Blood* 128(16):2083–88
66. Spindelboeck W, Schulz E, Uhl B, et al. 2017. Repeated fecal microbiota transplantations attenuate diarrhea and lead to sustained changes in the fecal microbiota in acute, refractory gastrointestinal graft-versus-host-disease. *Haematologica* 102(5):e210–13
67. Kaito S, Toya T, Yoshifuji K, et al. 2018. Fecal microbiota transplantation with frozen capsules for a patient with refractory acute gut graft-versus-host disease. *Blood Adv.* 2(22):3097–101
68. Qi X, Li X, Zhao Y, et al. 2018. Treating steroid refractory intestinal acute graft-versus-host disease with fecal microbiota transplantation: a pilot study. *Front. Immunol.* 9:2195
69. DeFilipp Z, Peled JU, Li S, et al. 2018. Third-party fecal microbiota transplantation following allo-HCT reconstitutes microbiome diversity. *Blood Adv.* 2(7):745–53
70. Taur Y, Coyte K, Schluter J, et al. 2018. Reconstitution of the gut microbiota of antibiotic-treated patients by autologous fecal microbiota transplant. *Sci. Transl. Med.* 10(460):eaap9489