Microb Health Dis 2023; 5: e877

# THE ROLE OF MICROBIOME IN CANCER TREATMENT: WHAT'S NEW IN 2023?

R. Jänsch, J. Bruns, M. Venerito

Department of Gastroenterology, Hepatology and Infectious Diseases, University Hospital Magdeburg, Medical Faculty, Otto-von-Guericke University, Magdeburg, Germany

*Corresponding Author:* Marino Venerito, MD; email: m.venerito@med.ovgu.de

**Abstract – *Objective:*** A growing body of evidence identifies the composition of the host gut microbiome as a predictor and target of therapeutic response in cancer patients.
***Materials and Methods:*** In this review, we searched PubMed to identify publications on the microbiome in cancer treatment published between March 2022 and March 2023.
***Results:*** The search identified six studies, four on the predictive role of the microbiome, and two interventional studies. Pre-therapeutic host gut microbiome composition analyzed by 16S rRNA gene sequencing was associated with therapeutic response in patients with various malignancies receiving CAR-T treatment or immune checkpoint inhibitors, while in a randomized, open-label phase 1 study, supplementation with the probiotic CBM588 containing Clostridium butyricum showed encouraging results in patients with metastatic renal cell cancer receiving nivolumab plus ipilimumab.
***Conclusions:*** The data presented highlight the opportunity for a personalized approach to cancer treatment based on the pre-therapeutic composition of the host gut microbiome and the targeting of gut microorganisms through dietary intervention.

**Keywords:** Microbiome, Cancer, check-point inhibitor, Immune therapy, Personalized medicine.

## INTRODUCTION

In recent decades the global incidence of cancer has increased, with an estimated 9,292,789 new cases and 958,133 cancer-related deaths in 2020, and a further increase is expected by 2040, mainly due to an aging population. The development of immune checkpoint inhibitors (ICIs) represents a new treatment option with a more favorable side effect profile for patients with many different types of cancer.

Microorganisms in the gut and other niches can modulate the efficacy of ICI therapies[1]. Over the past year, progress has been made in understanding the interplay between host-specific microorganisms and disease. Important new data suggest that knowledge of a patient's pre-therapeutic microbial composition and function may provide prognostic and predictive information. Furthermore, modulating or targeting a patient's microbial composition and function may have important therapeutic implications in the context of a multidisciplinary approach to cancer therapy. This systematic review summarizes recent advances in the role of the microbiome in cancer therapy published between March 2022 and March 2023.

## MATERIALS AND METHODS

A PubMed search was performed on April 3, 2023, for publications on the microbiome in cancer treatment during the previous year. Filters applied to the search were "custom date range" for articles published between March 2022 and March 2023, and article type clinical trial, randomized clinical trial, or meta-analysis. The Boolean operator "AND" was used to narrow the search results. The following search term combinations were used: "microbiome" [all fields] AND "cancer" [all fields]; AND "therapy" [all fields].

Studies were included if they were published in English in peer-reviewed journals, included cancer participants aged 16 years or older, and the microbiome was the focus of the study. Pediatric and animal studies were excluded. Study protocols and descriptive studies not related to cancer therapy were also excluded. All studies were screened by one reviewer (M.V.).

## RESULTS

The search strategy yielded 47 initial hits. Using the inclusion and exclusion criteria, 41 records were screened out at the title and abstract stage (16 did not include cancer patients, 16 did not focus on the microbiome, 4 were study protocols, 5 were descriptive studies – no interventional studies), leaving 6 reports to be assessed for eligibility at the full-text stage. This resulted in no further exclusions, and the six identified studies were included in the final analysis (Figure 1). These included four studies on the predictive role of the pre-therapeutic host gut microbiome and two interventional studies.

### Predictive Role of Microbiome for Cancer Treatment

In hematologic malignancies, chimeric antigen receptor T (CAR T)-cell therapy is an emerging strategy to target malignant cells. After receiving CAR T-cell therapy, patients may develop cytokine release syndrome (CRS) as a side effect, with symptoms ranging from mild to severe and life-threatening. To date, there is no reliable marker to predict the outcome of CAR T-cell therapy or the risk of developing CRS.

A recent study[2] investigated the role of the gut microbiome on both therapeutic response and risk of CRS in 99 patients with relapsed or refractory multiple myeloma (MM), treated with CAR T-cell therapy. With respect to safety, which was the primary endpoint of the study, 97% of

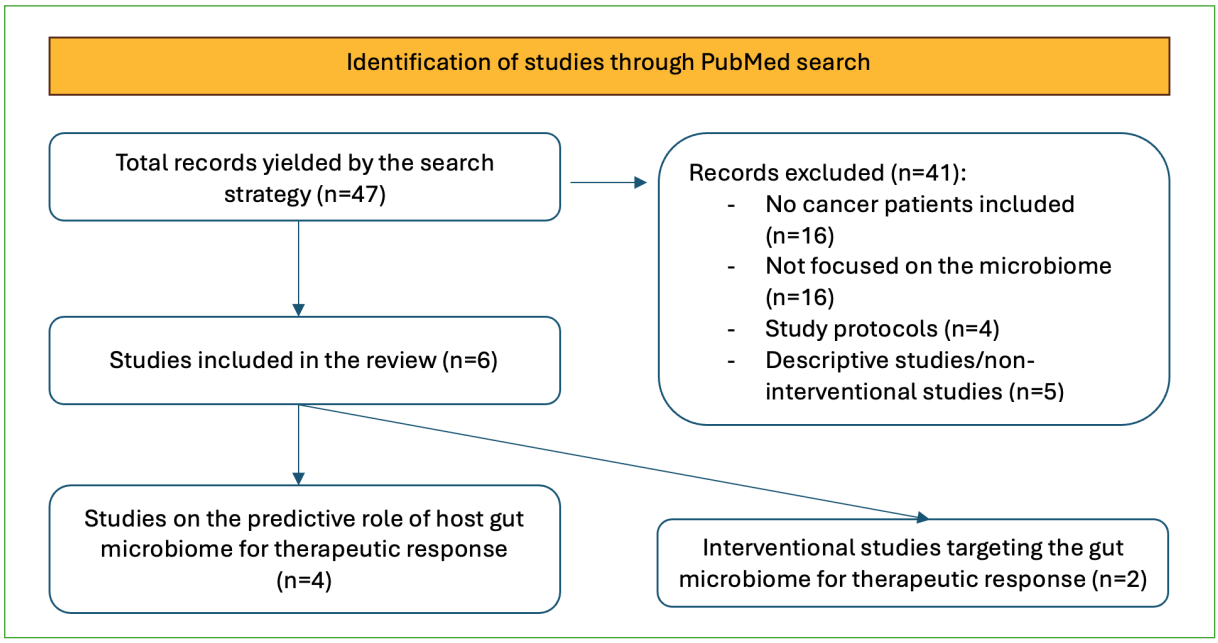


**Figure 1.** Flowchart of study identification and inclusion.

patients developed CRS ranging from grade 1 to 4, but all CRS resolved. In terms of short-term efficacy, nearly all patients responded to CAR T-cell therapy (96%) with complete (56%) or partial (40%) remission within one month.

Fecal samples from 81/99 patients with MM were available for 16S rRNA gene sequencing analysis of gut microbiome composition. Fecal samples were collected at five-time points (treatment stages): one before prior chemotherapy, one after chemotherapy before CAR T-cell infusion, and three times after CAR T-cell infusion. In general, the diversity of the microbiome decreased over the course of treatment. At the *phylum* level, the most abundant pre-treatment bacteria were *Firmicutes* and *Bacteroidetes*, with an increase in *Firmicutes* and a decrease in *Bacteroidetes* over the course of treatment. Further analysis at the taxonomic level from *phylum* to genus revealed an increase in *Enterococcus*, *Lactobacillus* and *Actinomycetes* and a decrease in *Bifidobacterium and Lachnospira* over time, with the greatest difference between stages for the genus *Enterococcus*. These results were further verified in another independent MM sample, as well as in two cohorts of patients with different hematological malignancies, namely acute lymphoblastic leukemia (B-ALL) and non-Hodgkin lymphoma (B-NHL).

To identify associations between the microbiome and treatment response, the authors compared the microbial composition or changes of patients with complete remission to those with partial remission. Interestingly, no differences in microbial composition were observed between groups at baseline. However, after CAR-T infusion, the phyla *Firmicutes* and *Bacteroidetes* and the orders Bacteroidales and Clostridiales were enriched in patients with complete remission. At the genus level, *Faecalibacterium*, *Roseburia*, and *Ruminococcus* were enriched after CAR-T, whereas *Prevotella*, *Collinsella*, *Bifidobacterium*, and *Sutterella* were enriched both before and after CAR-T cell infusion in patients with complete remission. *Suturella* was associated with longer progression-free survival. Thus, a functional role of the microbiome in the response of MM patients to CAR-T cell therapy can be hypothesized.

Regarding the association between the gut microbiome and CRS, the authors compared patients with severe CRS to those with mild CRS. While *Bifidobacterium* was enriched in patients with severe CRS both before and after the onset of CRS, *Leuconostoc* was enriched only during CRS. However, this observation could not be reproduced in an independent cohort of patients with MM. Furthermore, correlation network analyses revealed different correlations between leukocyte count or cytokine levels and commensals. For example, Lactobacillus correlated with IL-15 levels, whereas Clostridium correlated with higher lymphocyte counts.

Based on these findings, the development of novel biomarkers is warranted in order to optimize the management of patients with hematologic malignancies undergoing CAR T-cell therapy by predicting treatment outcome and CRS severity.

Another study[3] aimed to find common gut microbiome features predictive of response to ICI therapy for patients with different advanced-stage cancers regardless of tumor type. A meta-analysis[3] of 16S rRNA gut microbiome gene sequencing combining data from a discovery cohort of 16 patients with different advanced-stage cancers and those from three published cohorts of patients with melanoma treated by ICI found that certain gut bacterial taxa correlated with immunotherapy response status regardless of tumor type. In a second step, machine-learning models were used to investigate the association between patients' baseline gut microbiomes and clinical outcomes of immunotherapy. Responder microbiomes separated more clearly from non-responder microbiomes at the *phylum* level, suggesting that taxonomic rank may be an important variable to consider when searching for microbiome features that are consistently associated with response status. *Bacteroidetes* were more abundant in non-responders and *Firmicutes* were more abundant in responders. As a unique feature of this study, the use of the *selbal* analysis allowed for more complex community-based interactions to be uncovered, revealing differentially abundant groups of taxa (i.e., microbial signatures) that were not initially identified using univariate analyses.

In another retrospective study[4], 16S rRNA gut microbiome gene sequencing was performed on pre-treatment stool samples from patients with metastatic colorectal cancer (mCRC) and chemo-refractory non-small cell lung cancer (NSCLC) who received cetuximab (anti-EGFR antibody) and avelumab (anti-PD1-L antibody) in the CAVE-mCRC and CAVE-lung trials, respectively[4]. Patients in the CAVE-mCRC trial with longer progression-free survival (PFS) (9-24 months) had a higher abundance of the butyrate-producing bacteria *Aghatobacter M104/1* and *Blautia SR1/5* in their pre-treatment stool samples compared to patients with shorter PFS (2-6 months). This observation was confirmed in the validation cohort of patients in the CAVE-lung trial.

Both identified species produce butyrate, a metabolic modulator of innate and adaptive immunity under physiological and pathological conditions, which is associated with enhanced anti-PD-1 mAb efficacy and T cell infiltration in the tumor microenvironment in tumor-bearing mice. An evident limitation of this study is the small number of patients studied, and larger studies are warranted to confirm these interesting data.

Proton pump inhibitors (PPIs) are frequently prescribed in cancer patients. PPI can modulate gut microbiota and possibly alter responses to immune checkpoint inhibitor (ICI) therapy. In a meta-analysis of 41 retrospective studies, including 20,042 patients with advanced malignant tumors treated with ICIs, the potential association between PPI use and overall survival (OS) and/or PFS was analyzed[5]. In the included studies, patients were treated with ICIs alone or in combination with other anticancer drugs regardless of the therapeutic line. ICIs used included anti-PD-1, anti-PD-L1, and anti-CTLA-4. Overall, two studies showed a positive impact, 21 a negative impact, and 18 no significant effect of PPI. The evaluated endpoints were overall survival (OS) and progression-free survival (PFS). In this meta-analysis, the concomitant use of PPI was associated with impaired OS (HR=1.37; 95% CI, 1.23-1.52) and PFS (HR=1.28; 95% CI, 1.15-1.42) in patients treated with ICIs. In the subgroup analyses by primary tumor site, concomitant PPI use was associated with worse overall survival in patients with non-small cell lung cancer (NSCLC) and urothelial cancer, but not in patients with melanoma, renal cell cancer or hepatocellular carcinoma. The association between concomitant PPI use and impaired overall survival was also inconsistent across treatment lines, type of ICI and continents. No dose-dependent effect could be shown due to lack of data. The substantial heterogeneity observed was explained by the inclusion of different types of studies including *post hoc* analyses of prospective studies, retrospective studies and abstracts. The retrospective nature of the included studies is also responsible for a selection bias, which mainly explains the inconsistency of the results between subgroups. Thus, prospective studies are needed to determine the real impact of PPI use on survival outcomes in patients treated with ICIs.

## Interventional Studies

In preclinical studies, CBM588 containing *Clostridium butyricum* appeared to be able to enhance the growth of bifidobacteria. Additionally, in a retrospective study[6], CBM588 prolonged both PFS and OS in NSCLC patients receiving ICIs. In a randomized open-label phase 1 trial[6], 30 patients with metastatic renal cell cancer were randomized 2:1 to receive nivolumab plus ipilimumab with or without CBM588. Patients were assessed with imaging at 12-week intervals. Stool samples were collected from patients at baseline and after 12 weeks of treatment. The primary endpoint of the study, which was to determine the change in *Bifidobacterium* spp. collected from baseline to 12 weeks was not met as the abundance of *Bifidobacterium* spp. did not differ between the two groups at 12 weeks. However, the patients that received CBM588 showed a prolonged PFS with 12.7 *vs.* 2.5 months (Hazard ratio 0.15, 95% confidence interval 0.05-0.47, $p = 0.001$). Response rates were not significantly different but showed a trend toward better response in CBM588 patients with no change in toxicity. A major limitation is the small group size, and further studies are warranted to confirm this clinical observation and elucidate the mechanisms of action of CBM588 on the microbiome and response to ICI therapy.

A prospective randomized placebo-controlled trial[7] investigated whether postoperative probiotic administration can attenuate chemotherapy-induced gastrointestinal complications and gut microbiota dysbiosis in patients with CRC. One hundred patients with CRC who underwent surgery and were scheduled to receive chemotherapy (capecitabine and oxaliplatin, XELOX regimen) were randomized 1:1 to receive a probiotic combination or placebo from the third postoperative day until the end of the first course of chemotherapy. The combination of probiotics was administered *per os* in the form of a tablet containing *B. infants*, *L. acidophilus*, *E. faecalis* and *B. cereus* (live). Gastrointestinal complications such as nausea, heartburn, abdominal pain, abdominal distention, constipation and diarrhea were recorded. Fecal samples for 16S rRNA high-throughput sequencing and short chain fatty acid (SCFA) analysis were collected preoperatively and after the first cycle of postoperative chemotherapy. Chemotherapy significantly reduced the bacterial diversity of indexes of the gut microbiota, which in turn could be improved by probiotic supplementation. Chemotherapy also led to changes in the

composition of the microbiota, inducing a decrease in the *Firmicutes phylum* levels and an increase in the *Bacteroidetes*, *Proteobacteria*, and *Verrucomicrobia phylum* levels. Probiotic intake significantly reduced gastrointestinal complications, especially diarrhea, and restored taxa changes at both the *phylum* and the *genus* levels. In addition, probiotic supplementation increased the production of SCFAs.

An important limitation of this study is that fecal samples were not collected before the start of chemotherapy. Therefore, there may have been differences between the groups prior to probiotic supplementation. Another limitation is the lack of a group that did not receive chemotherapy. In fact, gastrointestinal symptoms, especially diarrhea, are a consequence of surgery rather than chemotherapy and should be treated as such. Causes of diarrhea after colon cancer surgery may include bile acid malabsorption and small intestinal bacterial overgrowth. Whether a probiotic supplementation can further improve properly diagnosed and treated gastrointestinal symptoms occurring following colon resection requires further study.

## CONCLUSIONS

Over the past year, progress has been made in understanding the role of the host gut microbiome in patients treated for cancer. The data presented highlight opportunities to target gut microorganisms through dietary intervention or other approaches to improve survival outcomes in patients treated for cancer. Additional prospective studies with larger sample sizes are underway to explore potential new microbiome-related therapeutic strategies in cancer. It is anticipated that in the next decade, pre-treatment gut microbiome profiling will become another pillar of decision-making for personalized cancer care, in addition to tumor and immune cell profiling, patient preferences and comorbidities, and prior therapies (Figure 2).

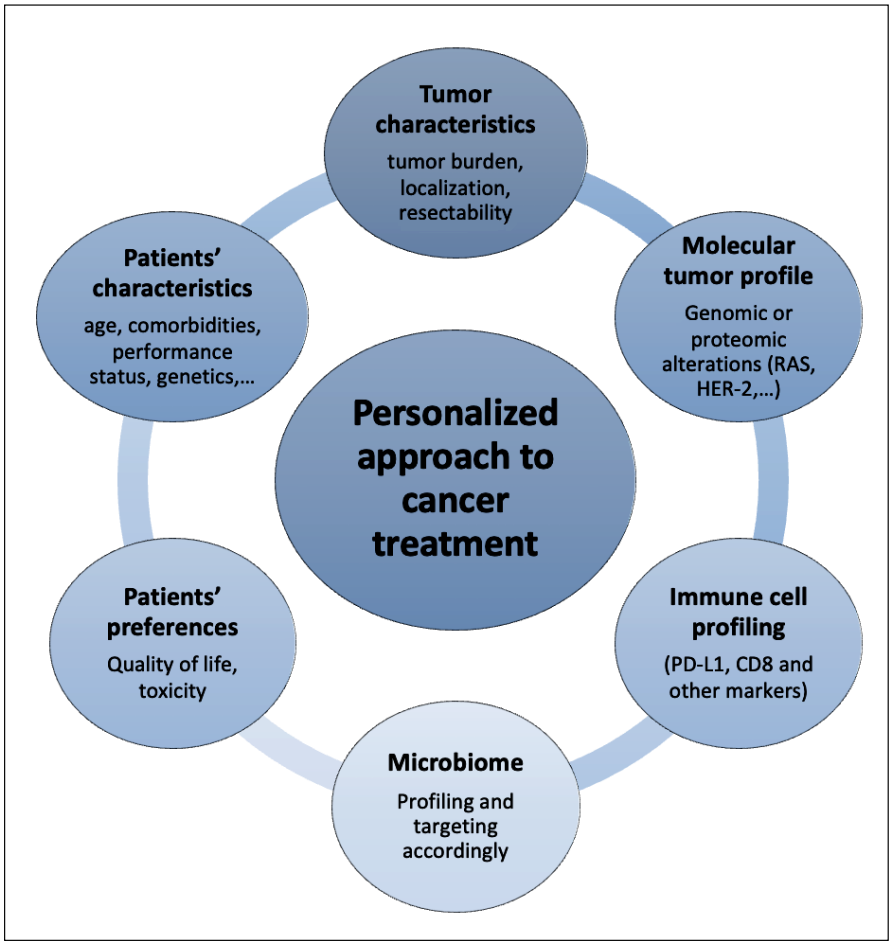


**Figure 2.** Personalized approach to cancer treatment: a forward-looking perspective considering microbiome profiling and microbiome-targeted interventions.

## Conflict of Interest

M.V. received honoraria for speaker, consultancy, and advisory role from Servier, Merck Serono, Bayer Vital, Roche, AstraZeneca, Ipsen, and is a member of the advisory boards of Ipsen, Lilly, Nordic Pharma, B.M.S., M.S.D., Eisai and Amgen. R.J. and J.B. have nothing to declare.

## Funding

None.

## Author's Contributions

Robert Jaensch: acquisition of the data, drafting of the article, Johannes Bruns: acquisition of the data, drafting of the article, Marino Venerito: conception and design of the study, acquisition of the data, drafting of the article, critical revision, supervision, final approval of the version.

## ORCID ID

Marino Venerito: https://orcid.org/0000-0001-8581-0974.

## Data Availability Statement

All data generated or analyzed during this study are included in this published article.

## REFERENCES

1. Park EM, Chelvanambi M, Bhutiani N, Kroemer G, Zitvogel L, Wargo JA. Targeting the gut and tumor microbiota in cancer. Nat Med 2022; 28: 690-703.
2. Hu Y, Li J, Ni F, Yang Z, Gui X, Bao Z, Zhao H, Wei G, Wang Y, Zhang M, Hong R, Wang L, Wu W, Mohty M, Nagler A, Chang AH, van den Brink MRM, Li MD, Huang H. CAR-T cell therapy-related cytokine release syndrome and therapeutic response is modulated by the gut microbiome in hematologic malignancies. Nat Commun 2022; 13: 5313.
3. Liang H, Jo JH, Zhang Z, MacGibeny MA, Han J, Proctor DM, Taylor ME, Che Y, Juneau P, Apolo AB, McCulloch JA, Davar D, Zarour HM, Dzutsev AK, Brownell I, Trinchieri G, Gulley JL, Kong HH. Predicting cancer immunotherapy response from gut microbiomes using machine learning models. Oncotarget 2022; 13: 876-889.
4. Martini G, Ciardiello D, Dallio M, Famiglietti V, Esposito L, Corte CMD, Napolitano S, Fasano M, Gravina AG, Romano M, Loguercio C, Federico A, Maiello E, Tuccillo C, Morgillo F, Troiani T, Di Maio M, Martinelli E, Ciardiello F. Gut microbiota correlates with antitumor activity in patients with mCRC and NSCLC treated with cetuximab plus avelumab. Int J Cancer 2022; 151: 473-480.
5. Lopes S, Pabst L, Dory A, Klotz M, Gourieux B, Michel B, Mascaux C. Do proton pump inhibitors alter the response to immune checkpoint inhibitors in cancer patients? A meta-analysis. Front Immunol 2023;14: 1070076.
6. Dizman N, Meza L, Bergerot P, Alcantara M, Dorff T, Lyou Y, Frankel P, Cui Y, Mira V, Llamas M, Hsu J, Zengin Z, Salgia N, Salgia S, Malhotra J, Chawla N, Chehrazi-Raffle A, Muddasani R, Gillece J, Reining L, Trent J, Takahashi M, Oka K, Higashi S, Kortylewski M, Highlander SK, Pal SK. Nivolumab plus ipilimumab with or without live bacterial supplementation in metastatic renal cell carcinoma: a randomized phase 1 trial. Nat Med 2022; 28: 704-712.
7. Huang F, Li S, Chen W, Han Y, Yao Y, Yang L, Li Q, Xiao Q, Wei J, Liu Z, Chen T, Deng X. Postoperative Probiotics Administration Attenuates Gastrointestinal Complications and Gut Microbiota Dysbiosis Caused by Chemotherapy in Colorectal Cancer Patients. Nutrients 2023; 15: 356.