The role of the human gut microbiome in non-small cell lung cancer response to immunotherapy treatment

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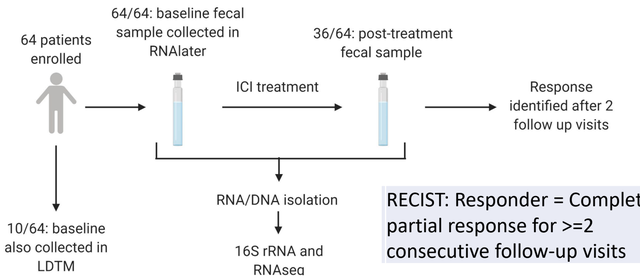
Introduction

Immune checkpoint blockade (ICB), a type of immunotherapy regimen, blocks the immune tolerance pathways overexpressed by tumor cells, allowing immune system components to remain activated against cancer cells. One type of ICB uses a monoclonal antibody directed against inhibitory receptor programmed death-1 (PD-1), expressed on the surface of immune cells, thereby blocking the PD-L1 signal present on tumor cells. Despite recent success in clinical trials using anti-PD-1, an estimated 40-60% of patients do not benefit from these therapies due to a lack of response. Recent studies have shown that the human gut microbial composition can determine whether a patient is a responder or non-responder to immunotherapy, and that fecal microbiota transplant (FMT) using responder stool can significantly improve PD-1-mediated anti-tumor response in mice. However, these studies have not identified common microbial signal shared by responding patients, and have not established a mechanistic explanation for how microbial-mediated response occurs.

Methods



Patient sample collection: In a partnership with Moffitt Cancer Center, we obtained pre-treatment (baseline) and post-treatment stool samples from 64 stage III/IV NSCLC patients undergoing ICB therapy. These patients were categorized as responders or non-responders to ICB using RECIST criteria.

16S rDNA sequencing: Fecal DNA was extracted from human or mouse fecal samples, and the 16S V1-3 region was amplified and sequenced using the Illumina MiSeq. The reads were processed using QIIME2, along with denoising, dereplicating, chimeras filtering and amplicon sequence variants (ASVs) generation using DADA2.

In vivo experiments: Patient stool was pooled by response phenotype (R: n=4, NR: n=6) and gavaged into wild type (WT) germ free (GF) mice housed in a bioexclusion system. 2 weeks post-colonization, mice were subcutaneously injected with 10^6 Lewis Lung Carcinoma cells followed by treatment with anti-PD-1. Tumor size was measured for 20 days, at which point tumors were excised. Tumor tissue was harvested and analyzed via flow cytometry and qPCR.

Figure 1: Responders have a different microbial community structure than non-responders at baseline.

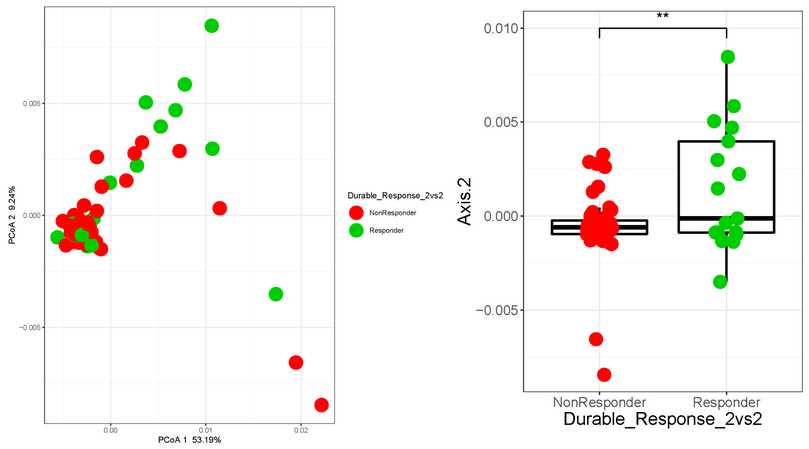


Figure 2: FMT from responder patients into mice reduces tumor growth following anti-PD-1 treatment

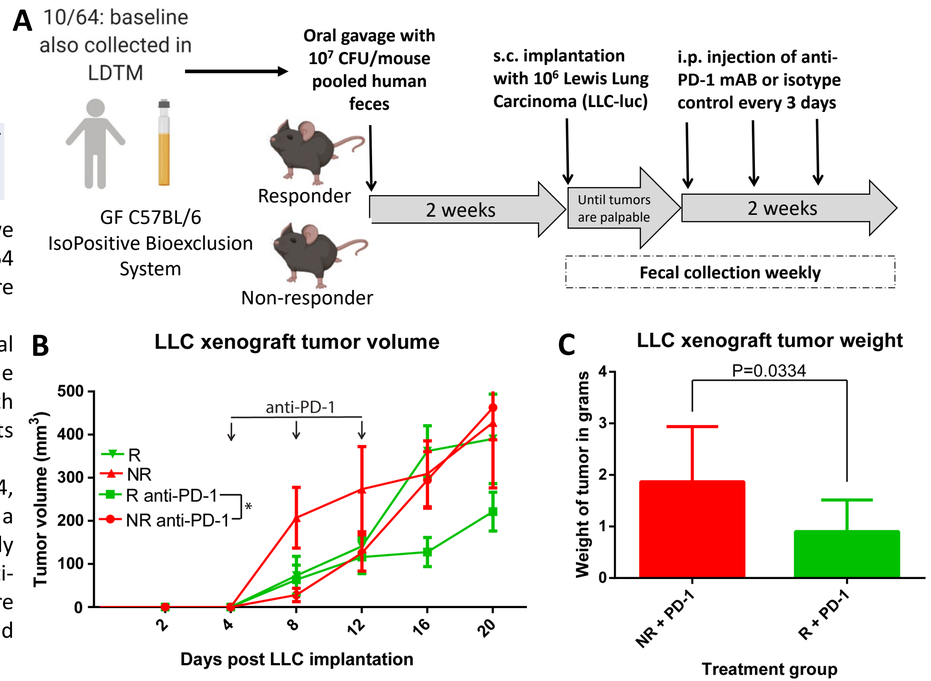
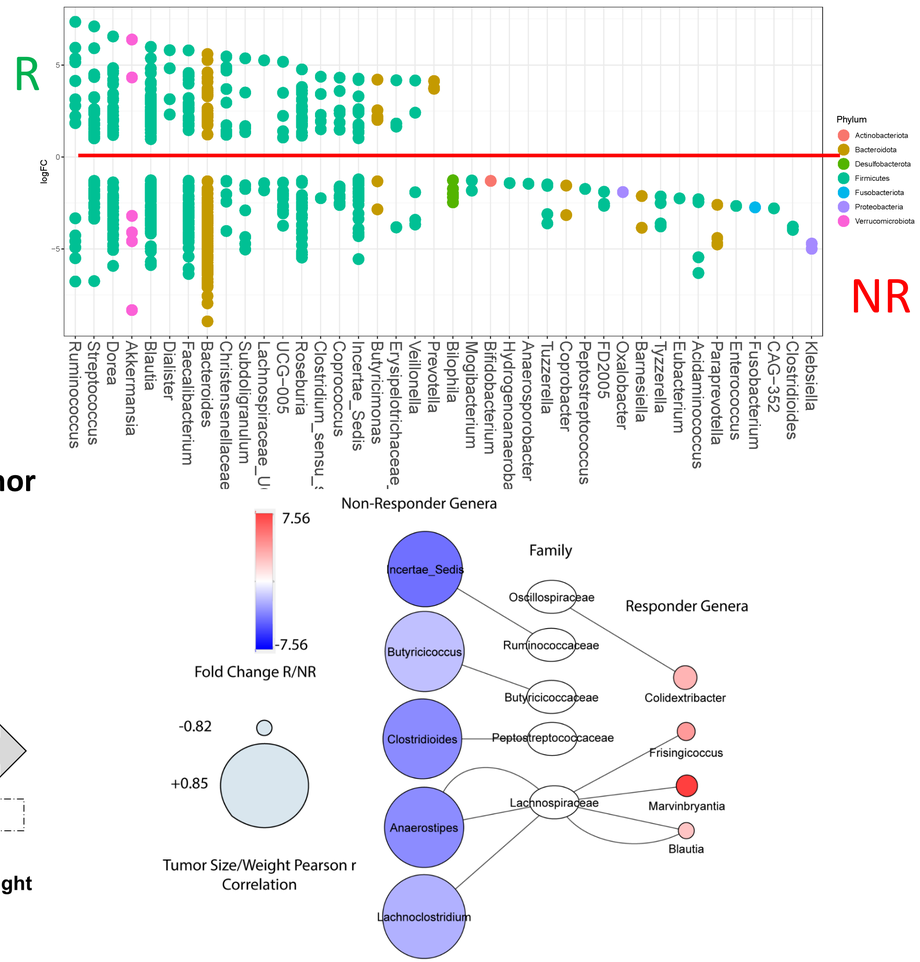


Figure 3: Specific taxa at the isolate level are associated with response and overlap between human and mice



Conclusions

The gut microbiota of NSCLC patients at baseline is different between those that respond and do not respond to immune checkpoint blockade. This response phenotype can be transferred to a germ free mouse using fecal transplant, and specific microbial taxa overlap between responders in humans and mice.

Acknowledgements

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