2839

DO NOT POST

Leveraging gut microbiota networks to impact tumor immunotherapy

Jaclyn Sceneay, Srimathi Srinivasan, Christopher Desjardins, Brian Weiner, Keith Halley, Meghna Bist, Kathleen Cieciuch, Kankana Bardhan, Edward O'Brian, Chris Ford, Jennifer Wortman, Matthew Henn, David Cook and Lata Jayaraman

SERES THERAPEUTICS, 200 Sidney St., Cambridge, MA, USA, www.serestherapeutics.com

Abstract

Background: The human gut microbiota forms a diverse, dynamic and complex ecosystem that modulates numerous host processes including metabolism, inflammation and cellular and humoral immune responses. Emerging data suggest that the gut microbiota of cancer patients may predict tumor response to immune checkpoint inhibitors (ICI) (Gopalakrishnan *et al*, 2018, Matson *et al*, 2018, Routy *et al,* 2018). To better understand how the microbiome may impact response to ICI and to evaluate the potential for therapeutic intervention, we have developed and validated robust tumor models using both conventional mice treated with antibiotics and germ-free (GF) mice.

Results:

(i) Our data show that in both models, mice lacking a diverse microbiome fail to mount an efficient anti-tumor immune response upon treatment with anti-PD-1.

(ii) Importantly for the first time, our data show that the bacterial spore fraction from healthy donor stool can increase CD8+ TILs in both conventional mice treated with antibiotics and GF mice in response to anti-PD-1, and thereby restore anti-tumor efficacy. This work has formed the basis for the development of SER-401, a first-in-class microbiome therapeutic currently in clinical trials in melanoma patients.

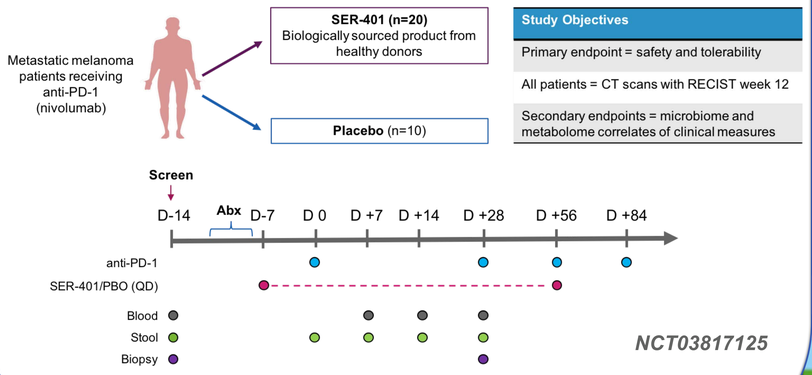
(iii) Functional characterization of the bacterial taxa in SER-401 can enable the construction of designed consortia of bacteria for the selection of a second generation clinical candidate.

(i) Preliminary studies suggest that bacterial ecologies can be successfully designed to elicit responses to anti-PD-1 in mouse tumor models.

We believe these data will provide insight into how microbiome drugs can be developed in the setting of immunotherapy to augment the efficacy of ICIs by altering the cancer-immune set point.

Clinical Trial: We are collaborating with the MD Anderson Cancer Center (MDACC) and the Parker Institute for Cancer Immunotherapy (PICI) on a randomized, placebo-controlled clinical study in patients with advanced metastatic melanoma. The trial is currently enrolling. This Phase 1b trial will evaluate the combination of an anti-PD-1 checkpoint inhibitor with adjunctive microbiome therapy, SER-401. Seres has developed SER-401, a purified suspension containing *Firmicute* spores derived from healthy human donors and formulated in capsules for oral administration.

Melanoma Checkpoint and Gut Microbiome Alteration with Microbiome Intervention (MCGRAW) – Ph Ib Clinical Study Schema



Acknowledgements: our grateful thanks to Dr Wargo & team (MDACC) and Dr Theresa LaVallee & team (PICI)

Development of SER-401

Lack of diversity of gut microbiota adversely impacts anti-PD-1 efficacy

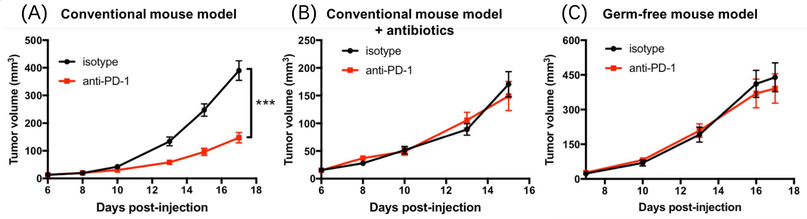


Figure 1. Tumor growth kinetics in C57Bl/6 mice bearing MC38 tumors and treated with anti-PD-1 on days 7, 10, 13 and 16. Shown in (A) conventional C57Bl/6 mice, (B) conventional mice treated with antibiotics (ampicillin, colistin, streptomycin) or (C) germ-free mice.

Anti-PD-1 efficacy is restored by spores derived from healthy donors

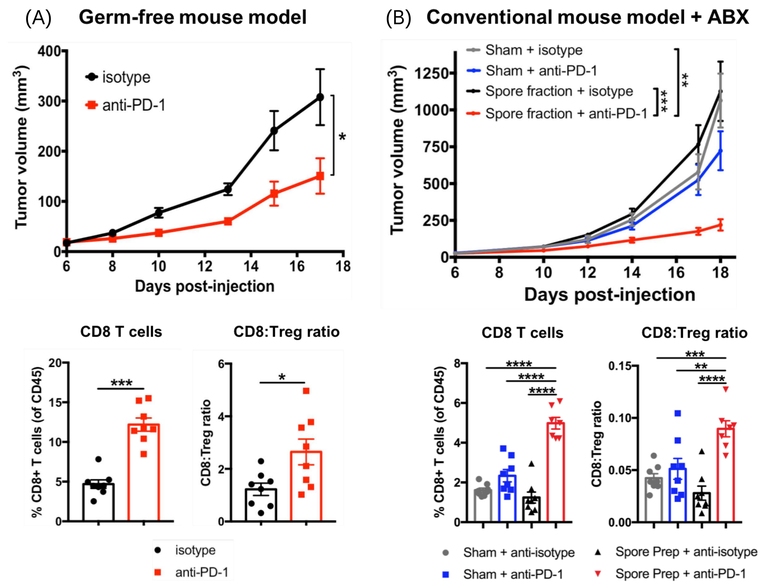


Figure 2. (A) Tumor growth kinetics in ex-germ-free mice bearing MC38 tumors and treated with anti-PD-1 on days 7, 10, 13 and 16. The spore fraction from healthy donor stool was administered 3 weeks prior to tumor cell injection. (B) Tumor growth kinetics in mice bearing MC38 tumors. Antibiotics were administered in the drinking water days -6 to 5. The spore fraction from heathy donor stool was administered on day 6, and on days 7, 10, 13 and 16 with anti-PD-1. T cell populations in MC38 tumors were assessed on day 17 by flow cytometry.

SER-401 Drug Substance restores anti-tumor efficacy

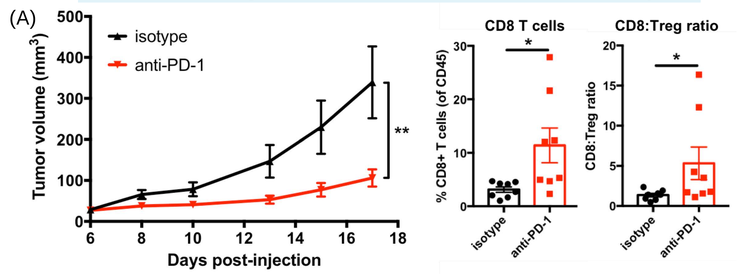


Figure 3. (A) Tumor growth kinetics in ex-germ-free mice bearing MC38 tumors and treated with anti-PD-1 on days 7, 10, 13 and 16. SER-401 Drug Substance was administered 3 weeks prior to tumor cell injection. Flow cytometry for T cell populations assessed on day 17.

Development of Designed Ecobiotics

Using functional and taxonomic data to inform construction of designed consortia

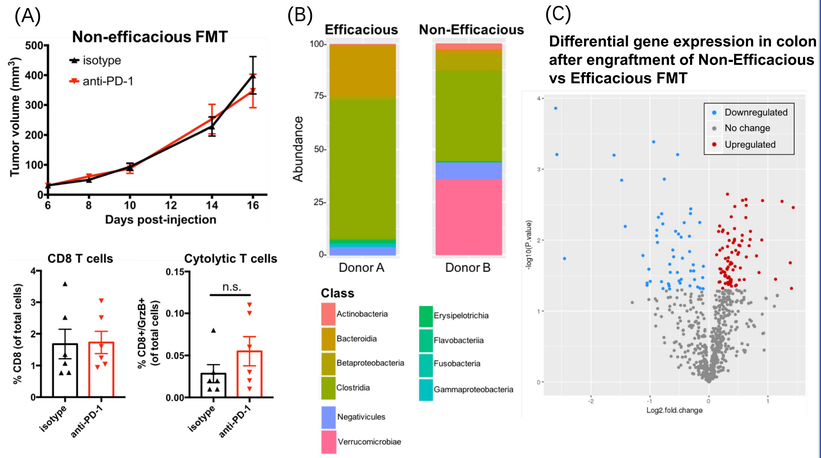


Figure 4. (A) Tumor growth kinetics in ex-germ-free mice bearing MC38 tumors and treated with anti-PD-1 on days 7, 10, 13 and 16 (n=10/group). FMT from donor stool was administered 3 weeks prior to tumor cell injection. Day 17 tumors were assessed for T cell populations by IHC. (B) OTU relative abundances (class level) of FMT inoculum material from efficacious and non-efficacious material as determined by 16SV4 sequencing analysis. (C) Germ-free mice were colonized with FMT material from (B). Colons were collected after 4 weeks and RNA analyzed using the Nanostring AutoImmune Profiling panel. Volcano plot shows differential gene expression as assessed by R Studio. Donor stool sample generously provided by Dr. Jonathan Peled (MSKCC).

Designed bacterial consortia shape the anti-tumor response to immunotherapy

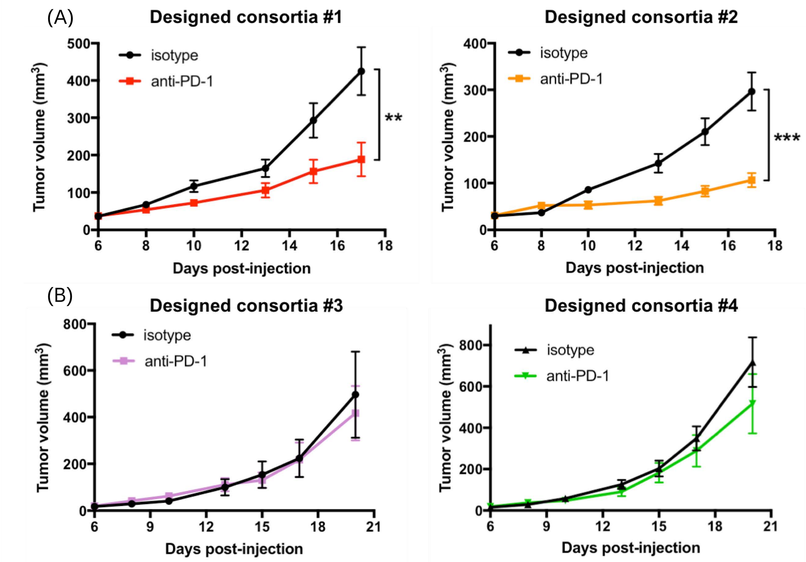


Figure 5. Tumor growth kinetics of ex-germ-free mice bearing MC38 tumors and treated with anti-PD-1 on days 7, 10, 13, 16. Designed consortia of (A) efficacious and (B) non-efficacious species were administered 3 weeks prior to tumor cell injection (on day 0).

n=8-10 mice per group for all studies

Statistical analysis was performed using two-tailed Student's T test or one-way ANOVA with p<0.05 considered significant (*p<0.05; **p<0.01, ***p<0.001, ****p<0.0001).