The Microbiome as a Biomarker and a Therapeutic Target

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Society for the Immunotherapy of Cancer (SITC)
Workshop on Nutrition, Metabolism and the Microbiome in Cancer Therapy
Session 1: The Impact of the Microbiome in Cancer Therapy
Washington, DC

November 8 2018

THE UNIVERSITY OF TEXAS
MD Anderson Cancer Center
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Disclosure information
SITC Workshop on Nutrition, Metabolism and the Microbiome in Cancer Therapy
Session 1: The Impact of the Microbiome on Cancer Therapy

The Microbiome as a Biomarker and a Therapeutic Target

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- I have the following financial relationships to disclose:

- Speaker's bureau: Imedex, Dava, Omniprex, Illumina, BMS, Merck, Gilead, MedImmune
- Advisory board member: Roche - Genentech, GSK, Novartis, Astra-Zeneca, Illiumina
- Clinical trial support: Roche - Genentech, GSK, BMS, Novartis

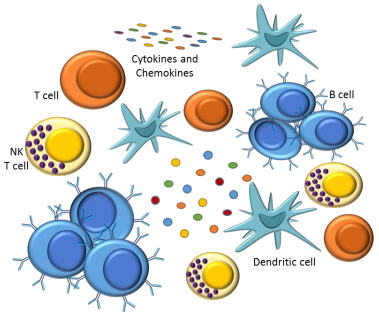
- I am a consultant and scientific advisor to Microbiome DX

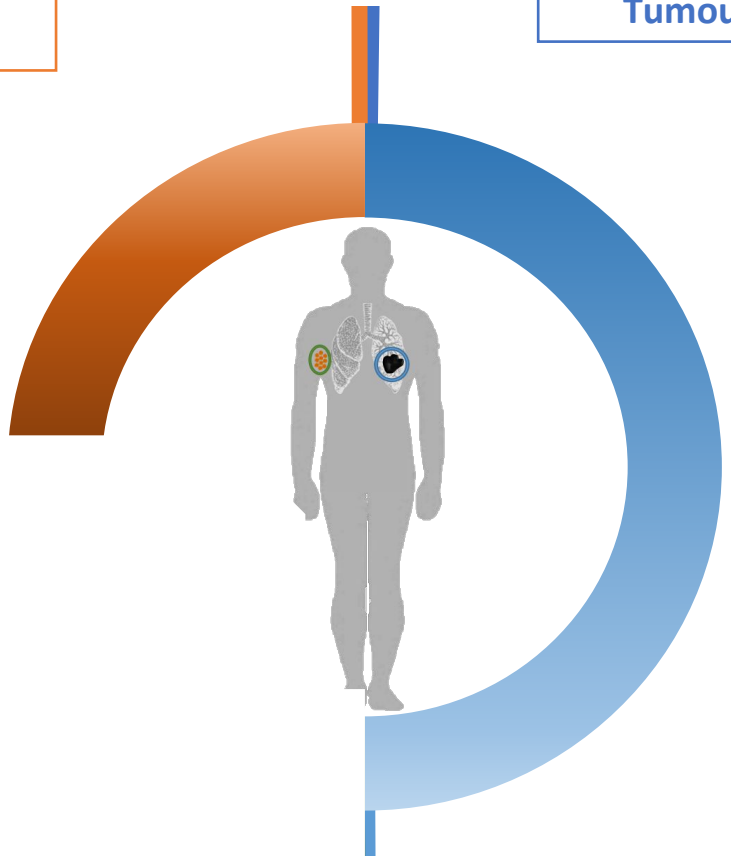
- *I am co –Inventor on patent submitted by The University of Texas MD Anderson Cancer Center to the US Patent and Trademark Office based on this work (Patent # PCT/US1/53717)*

There is a growing appreciation of the role of environmental factors in influencing cancer development and therapy

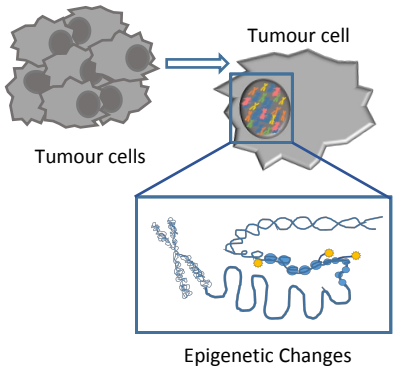
Systemic Immunity

Innate and Adaptive

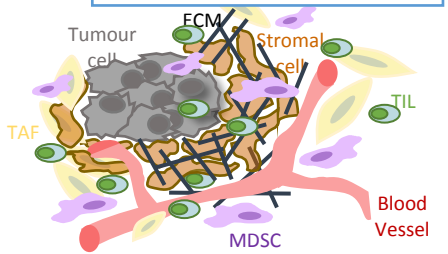




Tumour Genome and Epigenome

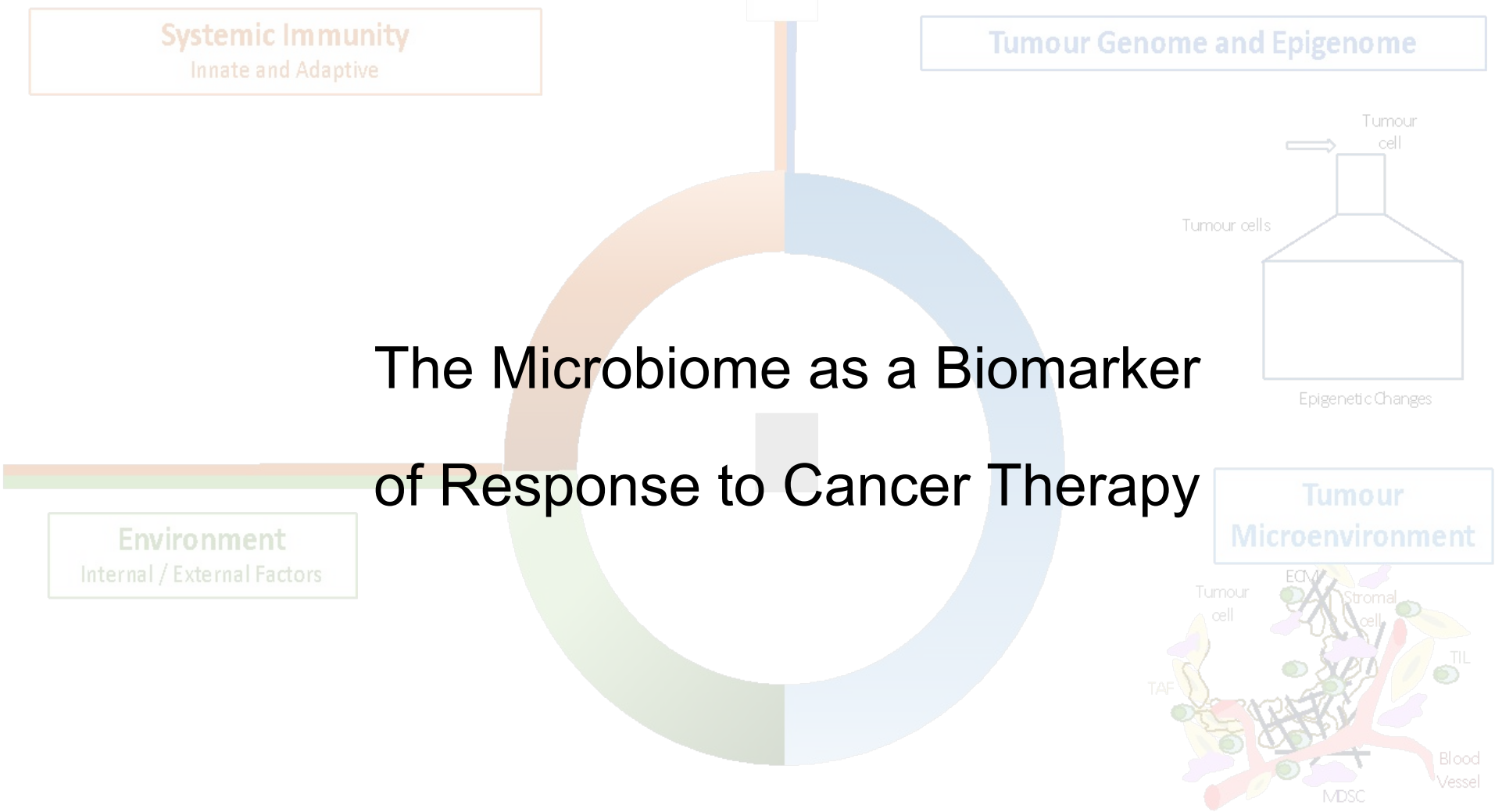


Tumour Microenvironment



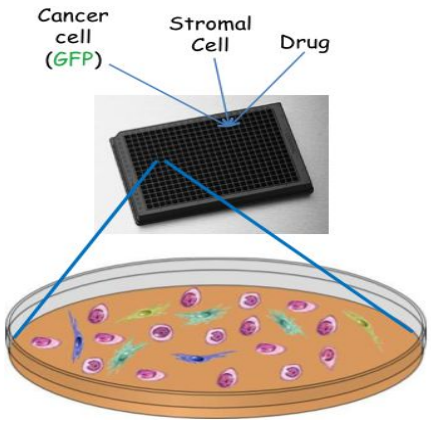
Cogdill, Andrews, Wargo - *British Journal of Cancer May 2017*

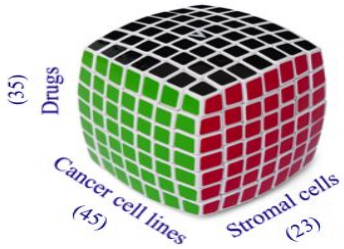
The Microbiome as a Biomarker of Response to Cancer Therapy

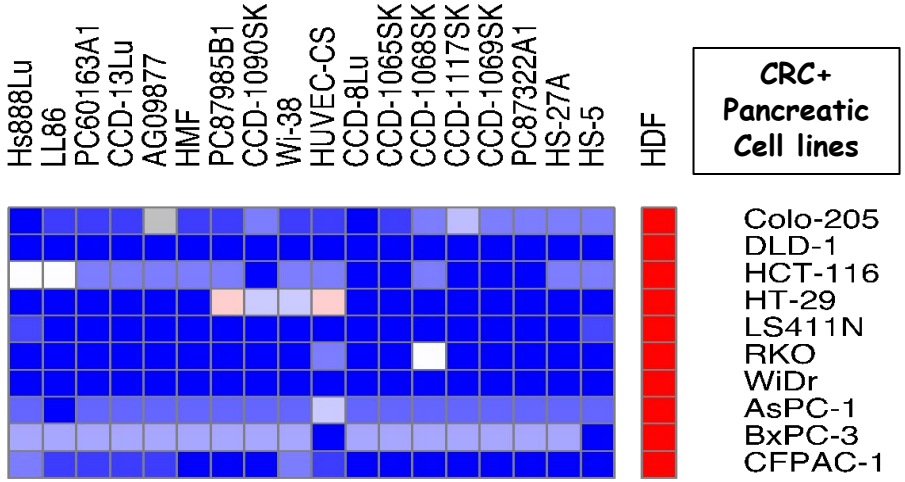


We studied the role of tumor stroma in resistance to therapy, and identified bacteria within cell lines derived from cancer patients that could confer resistance to therapy

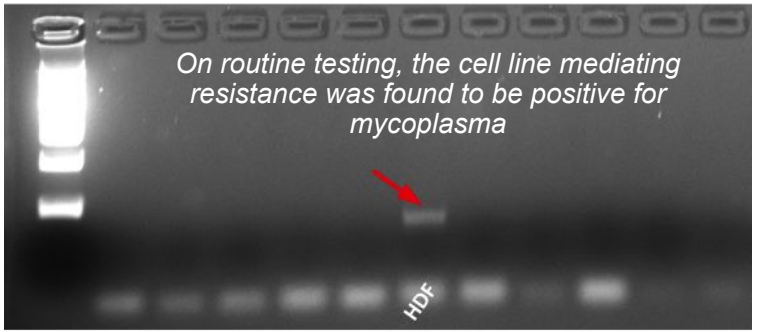
We worked with a team from MIT / the Broad to study resistance to chemotherapy in colorectal cancer and pancreatic cancer







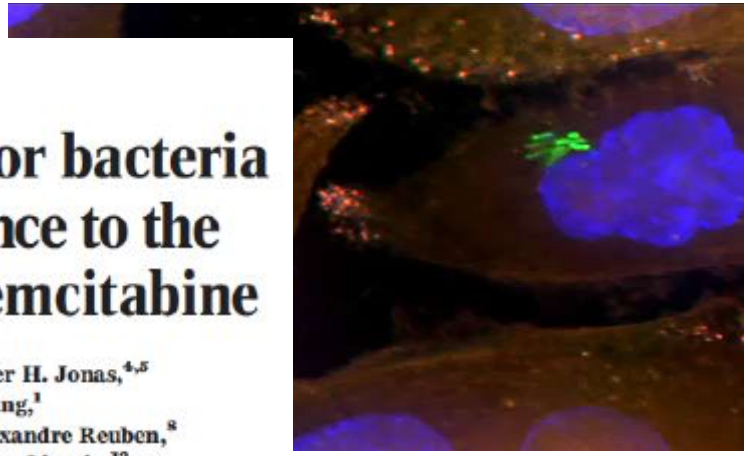
In these studies, one cell line rescued cancer cells from gemictabine



Mycoplasma is responsible for rescue from Gemcitabine:

- Eradication of mycoplamsa → no rescue
- Infection of another cell line → rescue
- WGS of HDF-pre-conditioned media → mycoplasma
- Bacteria were breaking down gemcitabine into inactive form

We validated these findings in patient samples, and showed that targeting co-targeting the bacteria and the cancer cells was associated with improved survival in mice

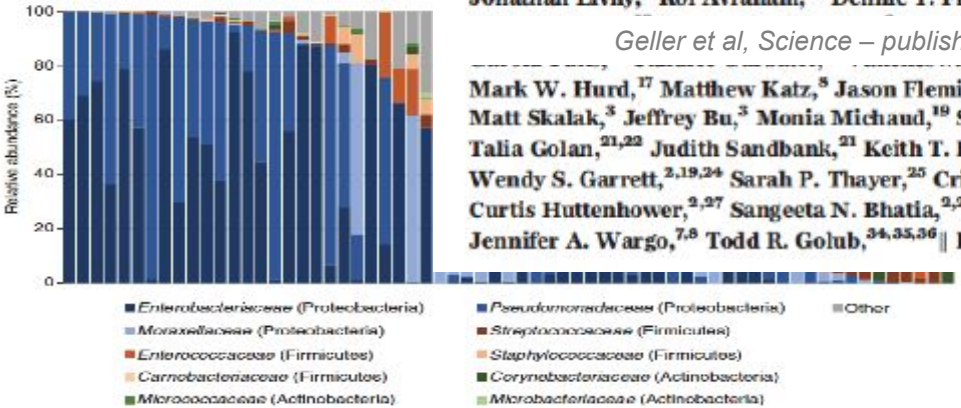


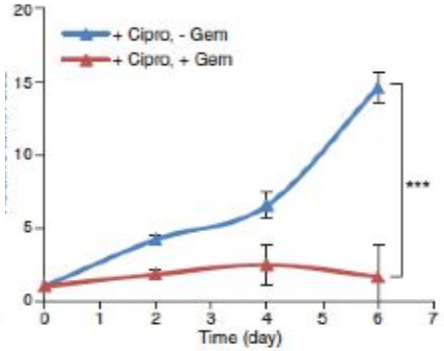
CANCER

Potential role of intratumor bacteria in mediating tumor resistance to the chemotherapeutic drug gemcitabine

Leore T. Geller,[1†] Michal Barzily-Rokni,[2*] Tal Danino,[3†] Oliver H. Jonas,[4,5] Noam Shental,[6] Deborah Nejman,[1] Nancy Gavert,[1] Yaara Zwang,[1] Zachary A. Cooper,[7,8‡] Kevin Shee,[2] Christoph A. Thaiss,[9] Alexandre Reuben,[8] Jonathan Livny,[2] Roi Avraham,[10] Dennie T. Frederick,[11] Matteo Ligorio,[12] Mark W. Hurd,[17] Matthew Katz,[8] Jason Fleming,[8] Anirban Maitra,[18] David A. Smith,[2] Matt Skalak,[3] Jeffrey Bu,[3] Monia Michaud,[19] Sunia A. Trauger,[13] Iris Barshack,[20,21] Talia Golan,[21,22] Judith Sandbank,[21] Keith T. Flaherty,[12] Anna Mandinova,[2,23] Wendy S. Garrett,[2,19,24] Sarah P. Thayer,[25] Cristina R. Ferrone,[26] Curtis Huttenhower,[2,27] Sangeeta N. Bhatia,[2,28,29,30,31,32,33] Dirk Gevers,[2§] Jennifer A. Wargo,[7,8] Todd R. Golub,[34,35,36]‖ Ravid Straussman[1]‖¶

Geller et al, Science – published September 15, 2017





With Ravid Straussman Todd Golub, Keith Flaherty, Dirk Gevers, Curtis Huttenhower et al

There is now a growing appreciation of the role of microbes in influencing cancer development, and evidence that microbes may impact therapeutic response

DOI: 10.1093/jnci/djs003
Advance Access publication on January 23, 2012.

Published by Oxford University Press 2012.

BRIEF COMMUNICATION

Fifteen-Year Effects of *Helicobacter pylori*, Garlic, and Vitamin Treatments on Gastric Cancer Incidence and Mortality

Jun-Ling Ma, Lian Zhang, Linda M. Brown, Ji-You Li, Lin Shi,
Wei-Dong Liu, Yuanreng Hu, Zhong ...
David Pee ...

New written informed consents were obtained for the extended follow-up phase from May 2, 2003, to August 1, 2010. Data from 3365 eligible participants ... 1–64 years ... both the

The NEW ENGLAND JOURNAL of MEDICINE

ORIGINAL ARTICLE

Immunoproliferative Small Intestinal Disease Associated with *Campylobacter jejuni*

LETTER

Proinflammatory CD4⁺CD45RB^hi Lymphocytes Promo... and Inte... in Apc^Min/+ Mice

Varada ...
Bruce H...

Adenoma-linked ...

doi:10.1038/nature11465

Foundational work to be presented by Dr. Christian Jobin at 11:30 am
""The interplay between diet, intestinal microbiota, and colorectal cancer"

Rese...

The In... Inflamm... carcinogenesis via Th2 cells

Atsuo Ochi,[1] Andrew H. Nguyen,[2] Andrea S. Bedrosian,[1] Harry M. Mushlin,[2] Saman Zarbakhsh,[1] Rocky Barilla,[1] Constantinos P. Zambirinis,[1] Nina C. Fallon,[1] Adeel Rehman,[1] Yuliya Pylayeva-Gupta,[3] Sana Badar,[1] Cristina H. Hajdu,[4] Alan B. Frey,[2] Dafna Bar-Sagi,[3] and George Miller[1,2]

Cell PRESS

...tumorigenesis

Grace Y. Chen,[1,3] Michael H. Shaw,[2,3] Gloria Redondo,[2,3] and Gabriel Núñez[2,3]

Division of Hematology and Oncology, ...

Intestinal Neoplasia in the *Apc^Min* Mouse: Independence from the Microbial and Natural Killer (*beige* Locus) Status[1]

William F. Dove,[2] Linda Clipson, Karen A. Gould,[3] Cindy Luongo,[4] David J. Marshall, Amy R. Moser,[5] Michael A. Newton, and Russell F. Jacoby

McArdle Laboratory for Cancer Research [L. C., A. R. M.] and Laboratory of Genetics [W. F. D., K. A. G., C. L.], University of Wisconsin, Madison, Wisconsin 53706; Department of Medicine, Division of Gastroenterology [D. J. M., R. F. J.] and Department of Biostatistics, Comprehensive Cancer Center [M. A. N.], University of Wisconsin, Madison, Wisconsin 53792

Promotion of Hepatocellular Carcinoma by the Intestinal Microbiota and TLR4

Dianne H. Dapito,[1,2,10] Ali Mencin,[3,10] Geum-Youn Gwak,[1,7,10] Jean-Philippe Pradere,[1,10] Myoung-Kuk Jang,[1] Ing... Hossein Khiabanian,[4,5] Adebowale Adeyemi,[3] Ramon Bataller,[6] Ja... an...

Chronic Active Hepatitis and Associated Liver Tumors in Mice Caused by a Persistent Bacterial Infection With a Novel *Helicobacter* Species

*Jerrold M. Ward, James G. Fox, Miriam R. Anver, Diana C. Haines, Cathi V. George, Michael J. Collins, Jr., Peter L. Gorelick, Kunio Nagashima, Matthew A. Gonda, Raymond V. Gilden, Joseph G. Tully, Robert J. Russell, Raoul E. Benveniste, Bruce J. Paster, Floyd E. Dewhirst, John C. Donovan, Lucy M. Anderson, Jerry M. Rice**

Some intra-tumoral microbes facilitate immune responses, while others hinder them

Potentiation of acute IFNγ responses by bacterial vesicles

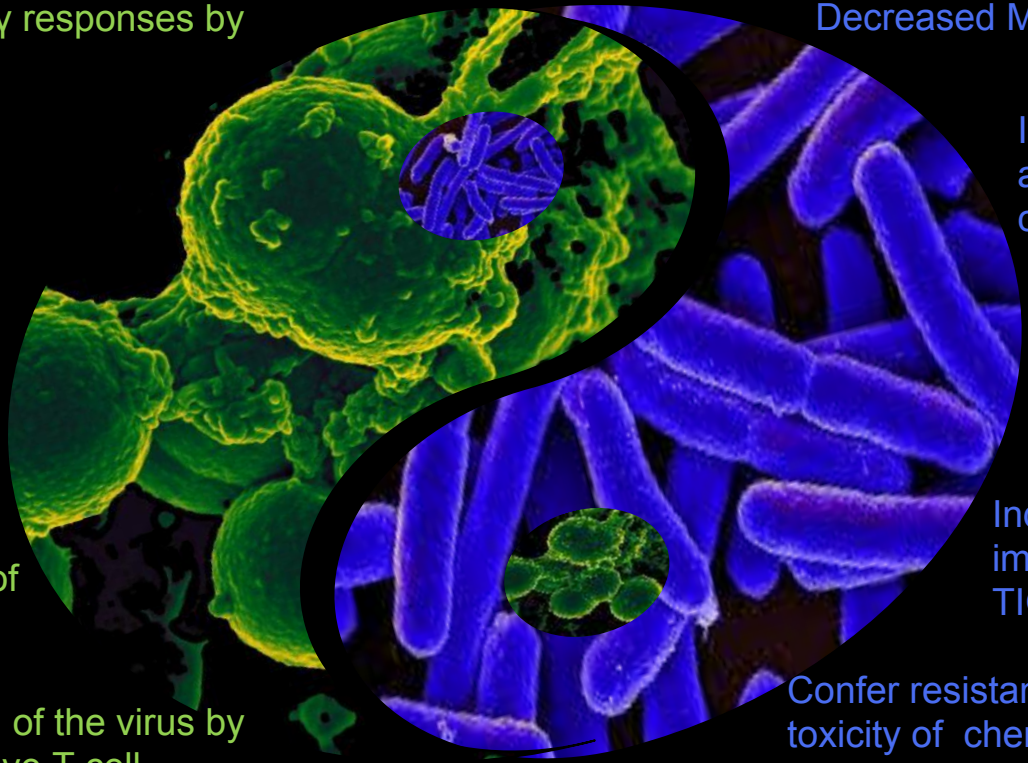
Direct engagement of innate immunity

Increased production of pro-inflammatory cytokines

Molecular mimicry

Increased expression of checkpoint molecules

Direct recognition of the virus by the TCR in adoptive T cell therapies and vaccines



Decreased MHC Class I expression

Increased production of anti-inflammatory cytokines

Toxins limit clonal expansion of lymphocytes

Induction of alternative immune checkpoints (eg TIGIT)

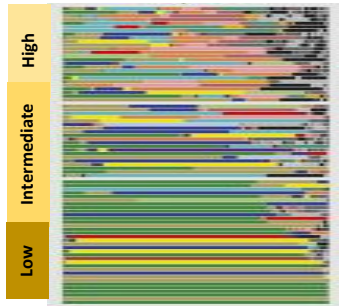
Confer resistance to and potentiate toxicity of chemotherapeutic agents

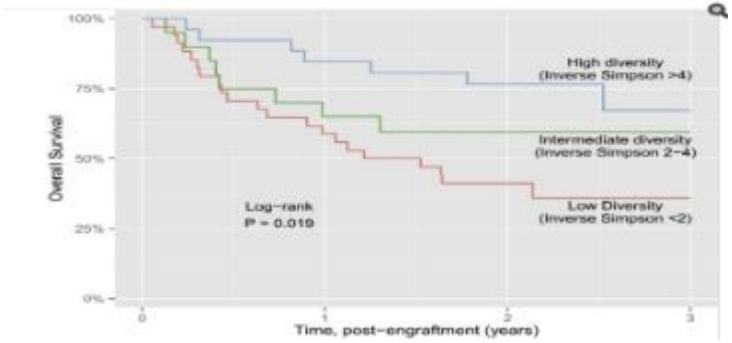
Beth Helmink MD PhD

Pierre Olivier-Gaudreau MD (PhD candidate)

During the course of our studies, we became aware of the outstanding work of others regarding influence of gut microbes on response to cancer immunotherapy

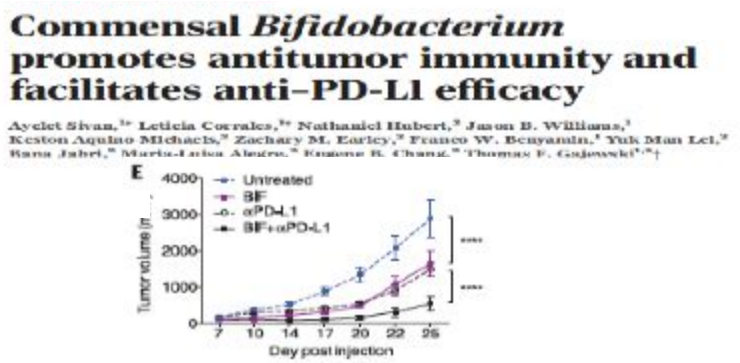
Diversity of the gut microbiome is associated with differential outcomes in the setting of stem cell transplant in patients with AML

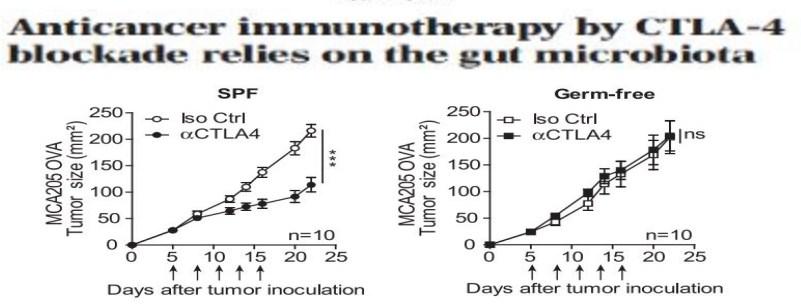




Taur…Pamer Blood 2014

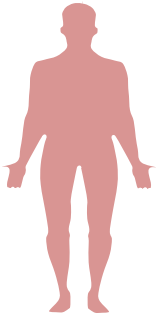
Composition of the gut microbiome is associated with differential responses to checkpoint blockade in murine models



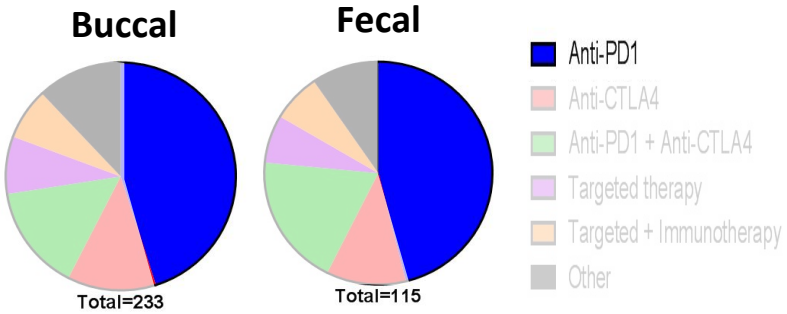


Sivan…Gajewski Science 2015, Vetizou…Zitvogel Science 2015

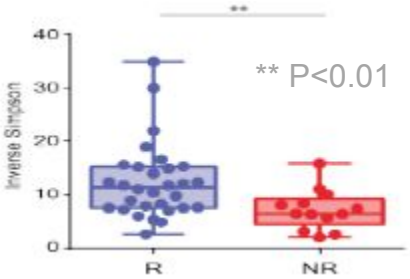
We studied oral and gut (fecal) microbiome in a large cohort of patients with metastatic melanoma going onto systemic therapy

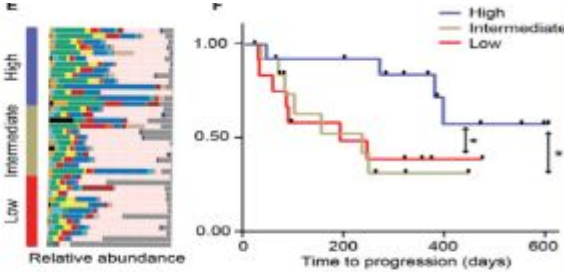


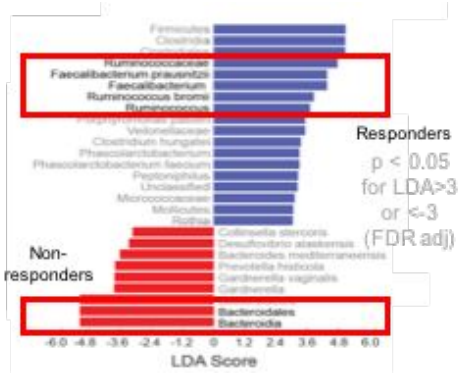
n = 233 patients



Microbiome sequencing & immune profiling was performed







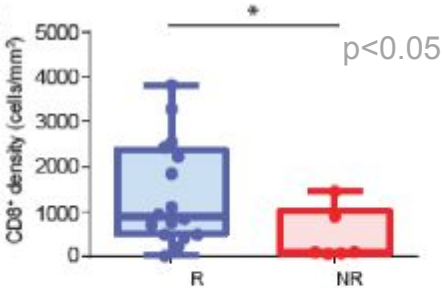
Responders to anti-PD-1 had a higher diversity of gut bacteria associated with prolonged PFS (along with additional compositional differences)

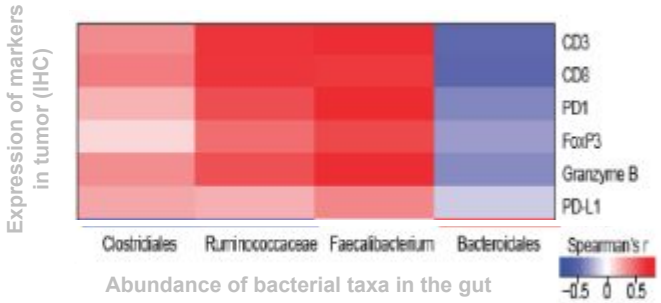
Deepak Gopalakrishnan PhD

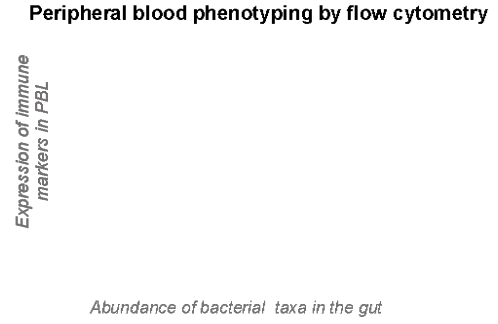
Gopalakrishnan et al, Science 2018

Christine Spencer PhD

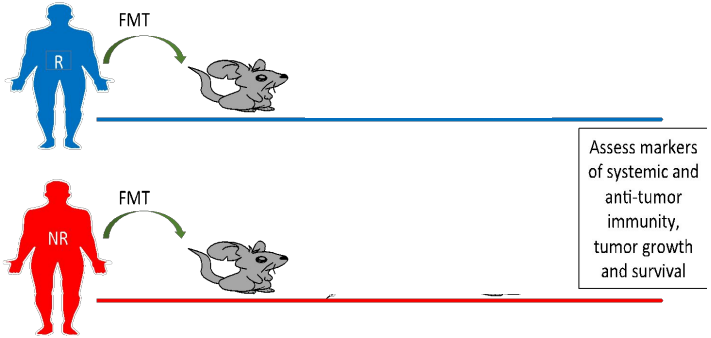
Importantly, “favorable” signatures in the gut microbiome were associated with enhanced immune responses in the tumor microenvironment

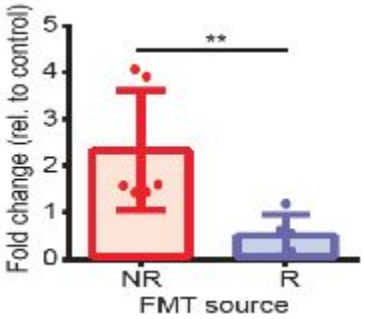


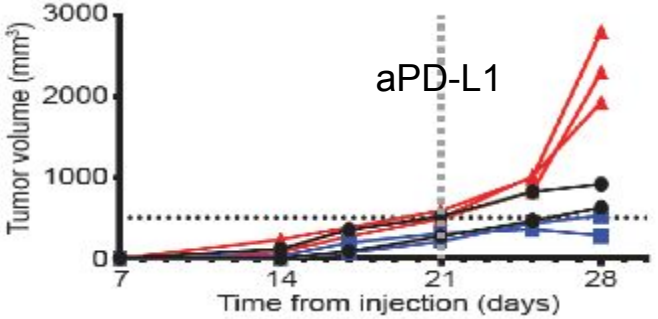




And mechanistic studies in germ free mice showed that fecal transplant could recapitulate the phenotype







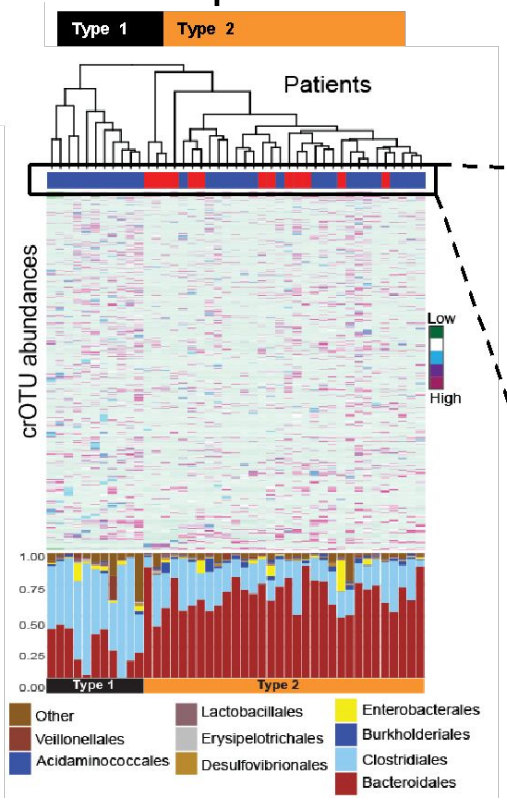
Mechanistic insights suggest that this is mediated both at the level of the gut and mesenteric lymph node, and also potentially via metabolites produced by gut microbes potentially mediating distant effects

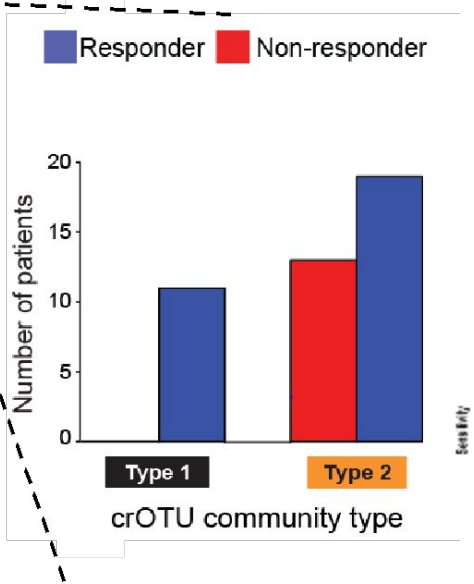
Alexandre Reuben PhD

Gopalakrishnan et al, Science 2018

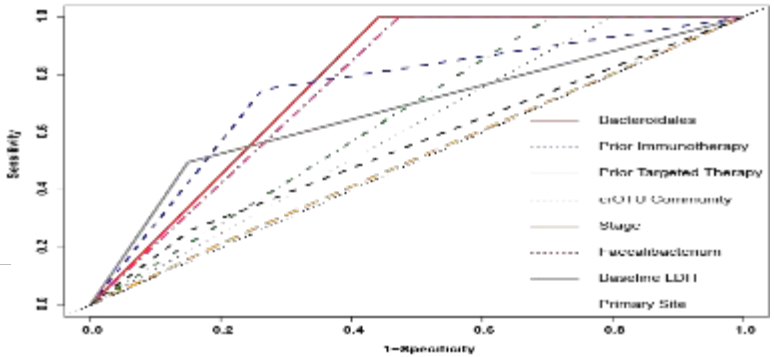
Luigi Nezi PhD

In our cohort, we identified a gut microbiome “signature” with a high likelihood of response to anti-PD-1 (with subsequent validation in a larger cohort)





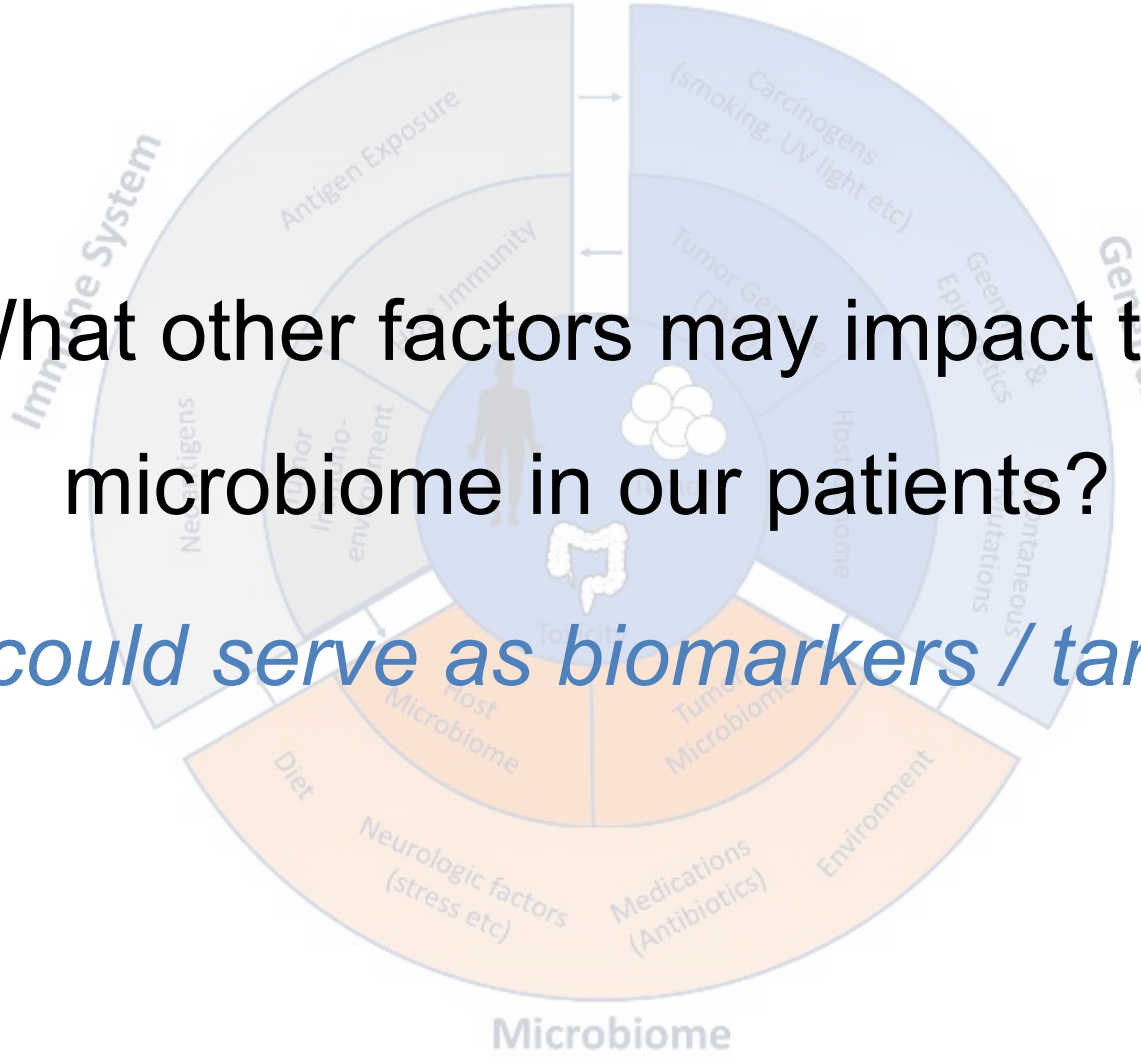
Suggesting that the gut microbiome could be used as a biomarker of response to immune checkpoint blockade, with patients with a “type I” signature more likely to respond



Peter Prieto MD MPH

Gopalakrishnan et al, Science 2018

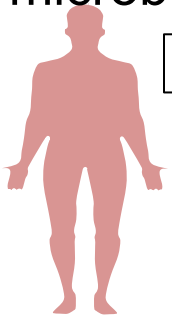
Alex Cogdill MS (PhD candidate)



What other factors may impact the microbiome in our patients?

(that could serve as biomarkers / targets?)

In our cohort, we also studied the influence of diet and lifestyle factors on the microbiome and response to therapy, and studied this with the type I signature



Diet & lifestyle survey

REDCap

Initial oral & gut microbiome sampling

Tumor biopsy

Start of therapy

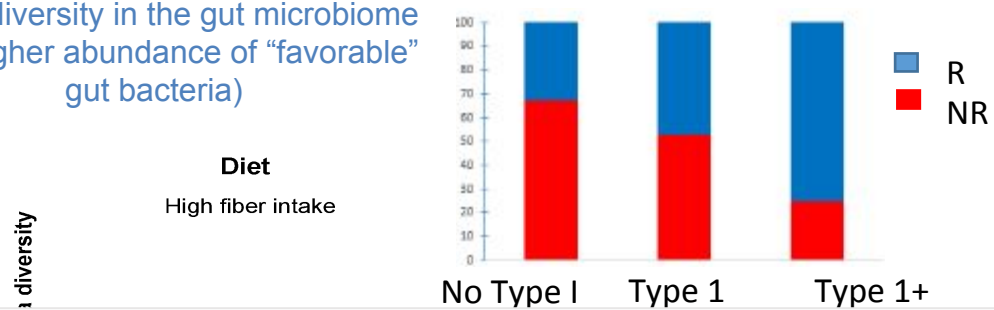
Clinical assessment & restaging

Repeat oral & gut microbiome sampling

Christine Spencer PhD

Lorenzo Cohen PhD

Patients with a high fiber diet had higher diversity in the gut microbiome (with higher abundance of "favorable" gut bacteria)



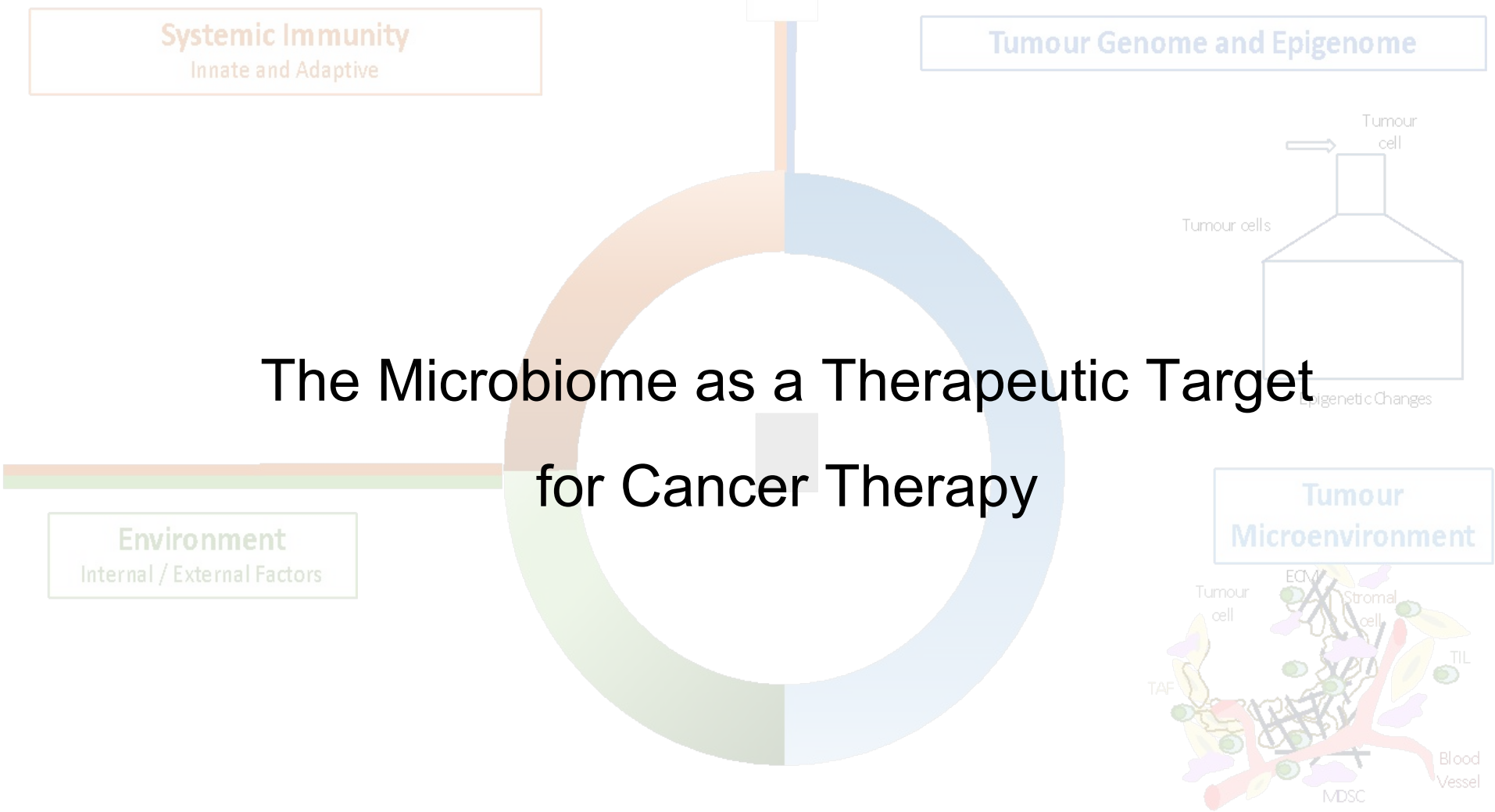
Factor	OR (95% CI)
No signature	1.00 (ref)
Type I Signature -Low Fiber	2.85 (0.78-10.47)
Type I Signature - High Fiber	9.00 (1.27-63.9)
P_{trend}	0.02

More data to be presented by Dr. Carrie Daniel-MacDougall
"Harnessing diet and the microbiome for cancer patients and survivors" 10:30 am today
and by Dr. Deepak Gopalakrishnan – Poster session (P505)

Carrie Daniel MacDougall PhD

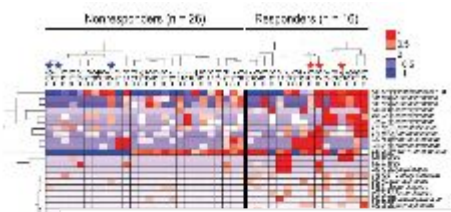
Jen McQuade MD

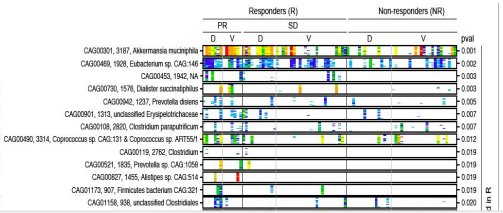
The Microbiome as a Therapeutic Target for Cancer Therapy

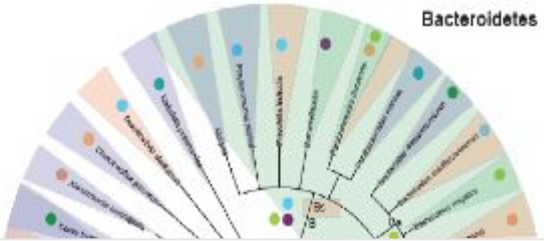


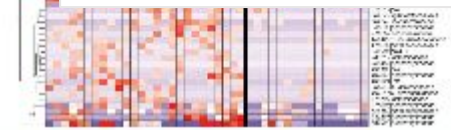
Numerous studies in human cohorts now support a link between the microbiome and response and toxicity to cancer therapy

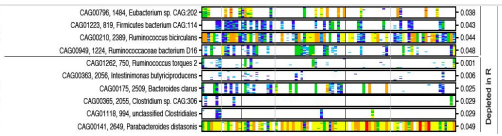
Studies in patients with melanoma, RCC, and NSCLC demonstrate differential "signatures" in R vs NR to ICB

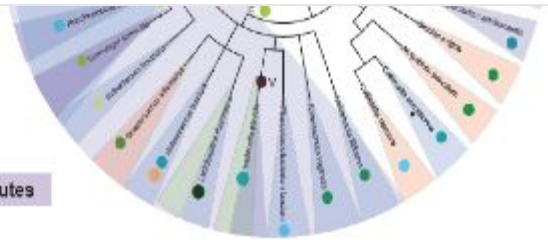




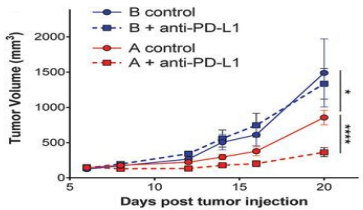


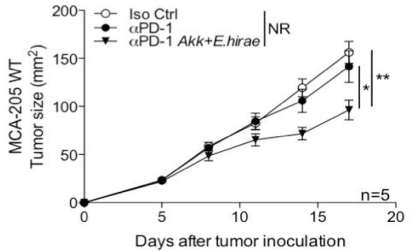






Together, these studies support the notion that we could potentially modulate the microbiota to improve responses and / or abrogate toxicity

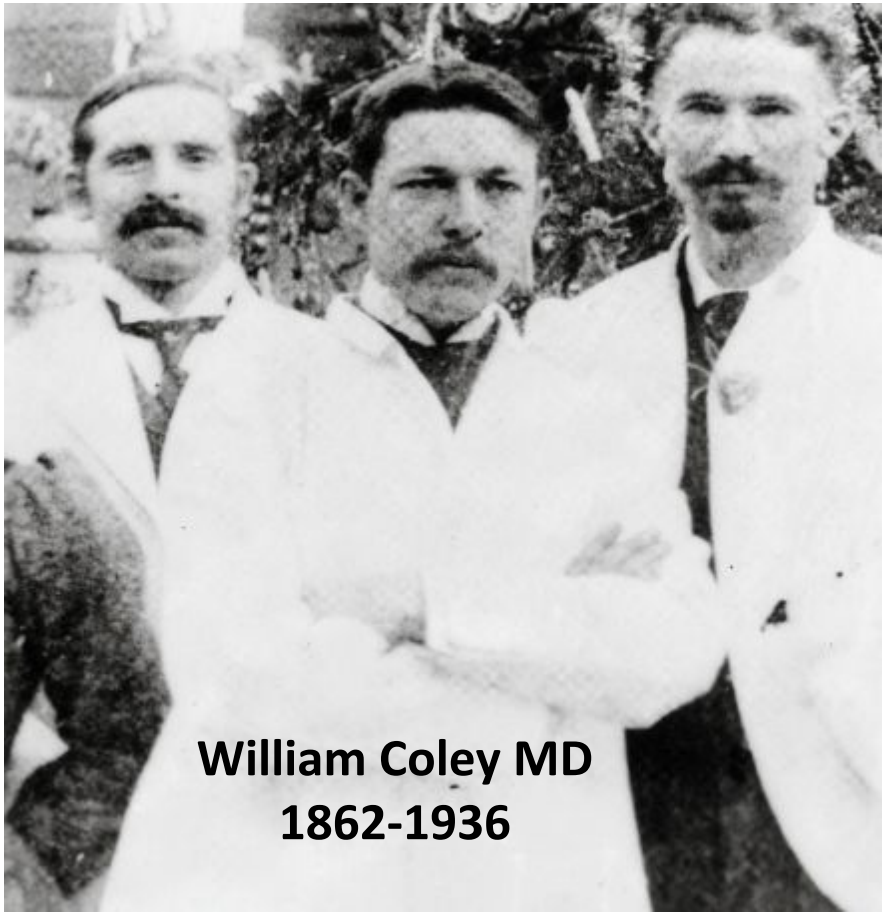




Matson et al, Routy et al, Science 2018

Gopalakrishnan, Helmink et al, Cancer Cell 2018

This includes efforts to target intra-tumoral microbes

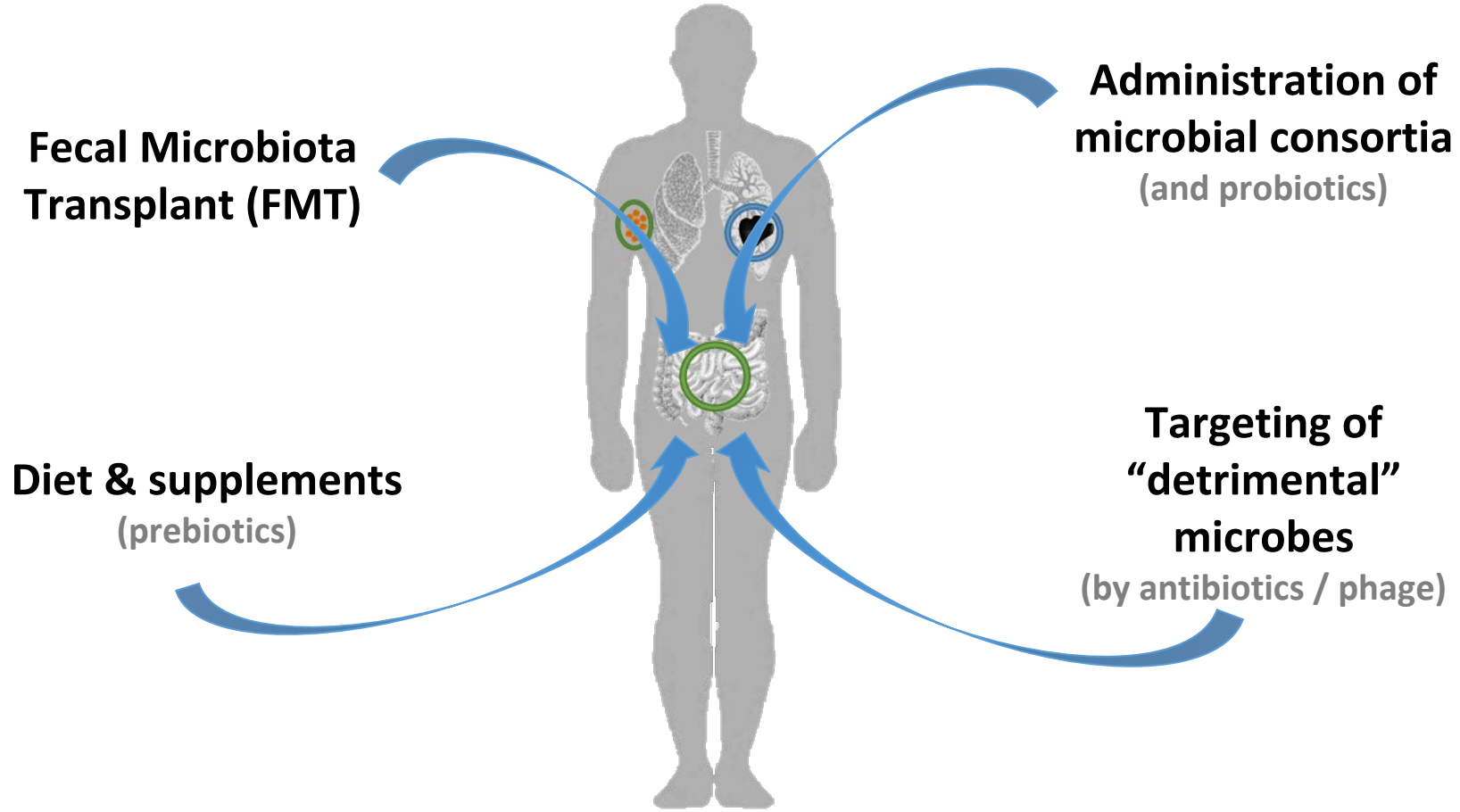


A. Targeting the intra-tumoral microbiome

Type of therapy	Target	References
Antibiotics		
Ciprofloxacin	*Gammaproteobacteria*	[49]
Metronidazole	*Fusobacterium nucleatum*	[127]
Targeted agents		
β-glucuronidase inhibitors	β-glucuronidase enzyme	[134]
Immunotherapy		
Adoptive T Cell therapy	EBV	[70]
	HPV	[39]
	CMV	[39]
	MCPyV	[39]
Vaccines	HPV	[134]
Anti PD-1 / PD-L1 immune checkpoint blockade	MCPyV HBV / HVC	[62]

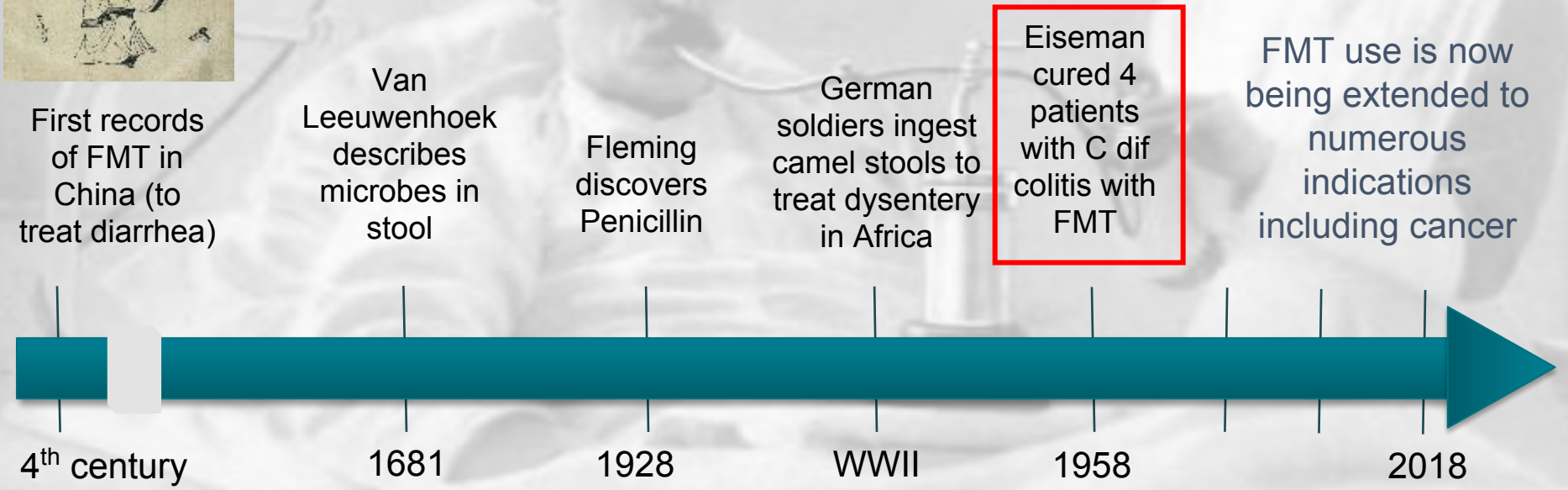
Cogdill et al, *Trends in Immunology* 2018

This also includes efforts targeting gut microbiota (via several different strategies)

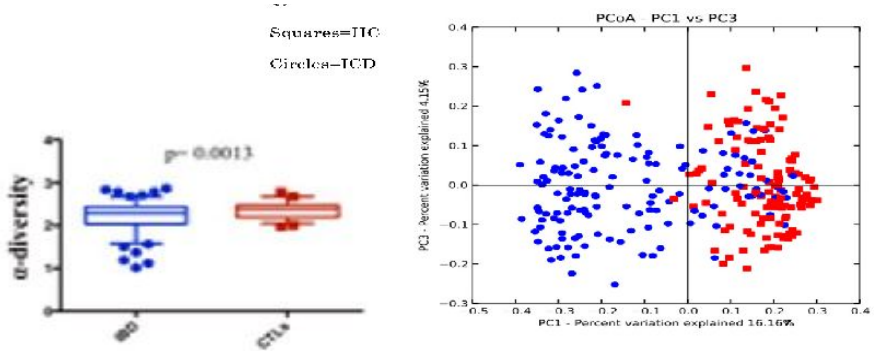


Fecal Microbiota Transplant (FMT) has been used to treat disease for centuries

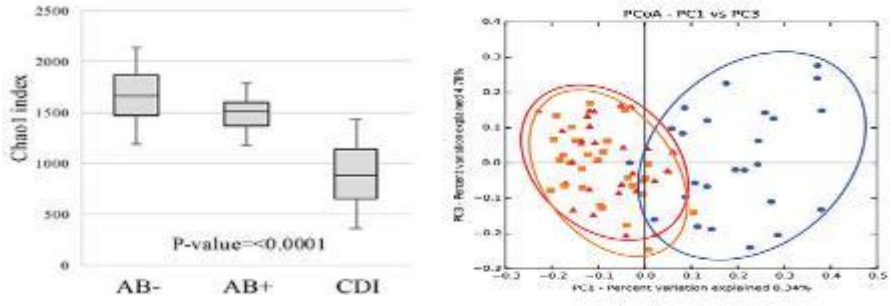
FMT is a logical (and likely necessary) first step in understanding how best to modulate the gut microbiome to enhance responses to therapy (and to abrogate toxicity)



Fecal Microbiota Transplant (FMT) has been successfully used in the treatment of diseases associated with dysbiosis of the gut microbiome *(e.g. IBD, CDI)*

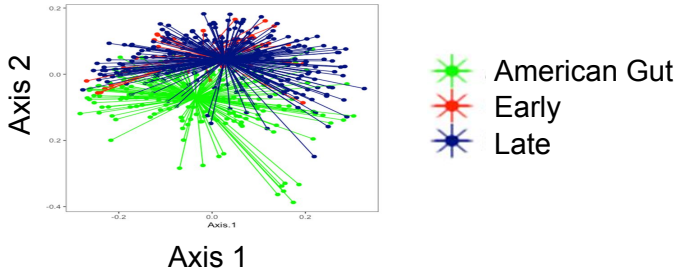


Walters, Xu & Knight FEBS Letters 2017



Milani et. Al. Scientific Reports, 2016

Patients with IBD and CDI have significant dysbiosis compared to healthy controls, and treatment with healthy donor FMT has been successful in treating these conditions



We have data demonstrating that the gut microbiota of melanoma patients is distinct from healthy individuals, suggesting that a relative dysbiosis may be at play here as well

Spencer, Goplakrishnan, McQuade et al, confidential unpublished data * PLEASE DO NOT POST *

Several trials are now underway involving strategies to modulate the microbiome in combination with immune checkpoint blockade (using FMT from CR donors)

Trial number	Patient population	n	Intervention	Outcome
NCT03353402	Metastatic melanoma patients resistant to CPI	40	Single arm: FMT from CPI responders via colonoscopy followed by stool capsules	Engraftment and safety; immune profile change
NCT03341143	Metastatic melanoma patients resistant to CPI	20	Single arm: FMT from anti-PD1 responders via colonoscopy + anti-PD1	ORR; immune profile change

*McQuade et al, manuscript under review * **DO NOT POST***

However screening of CR donors via sequencing should be performed- as not all CRs have a "favorable" gut microbiome (nor do all "healthy" donors!)

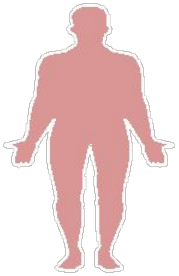
More data to be presented by Dr. Beth Helmink
"Variation of the gut microbiome in CRs to immune checkpoint blockade and healthy donors – implications for clinical trial design" – Poster session (P572)

Beth Helmink MD PhD

Deepak Gopalakrishnan PhD

PICI-0014: A randomized trial to evaluate the impact of gut microbiome modulation in patients going on to treatment with immune checkpoint blockade (MCGRAW)

PICI0014



Patients with metastatic cancer (melanoma) going onto immune checkpoint blockade (anti-PD-1)

Arm A
FMT from complete responders (CRs) to aPD-1
(n=20)

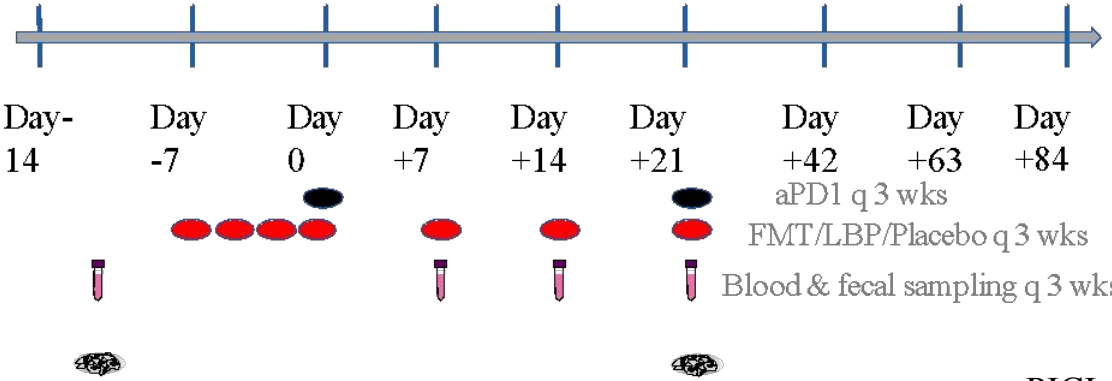
Arm B
Ser-401 (Live bacterial product – spore prep with Type 1 signature)
(n=20)

Arm C
Placebo
(n=20)

All patients = CT scans with RECIST week 12

Primary endpoint = safety and tolerability

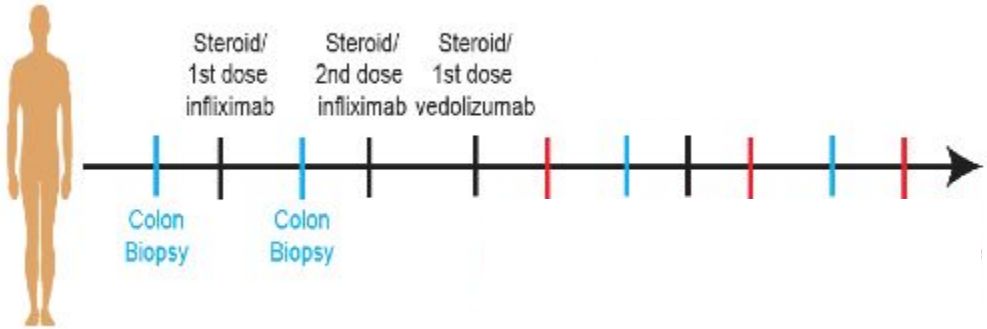
Secondary endpoints: engraftment, response and correlative studies (immune correlates in blood and tumor, metabolites)



MDACC: Tawbi, Glitza, Burton

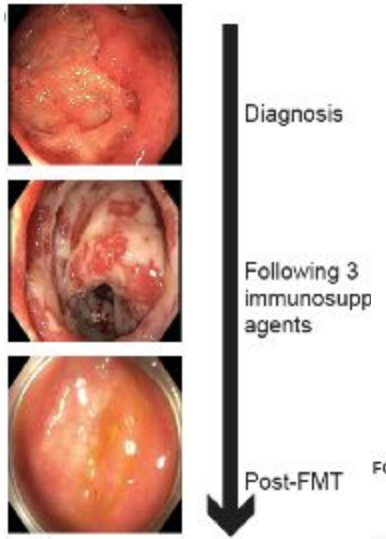
PICI: Ibrahim & LaValle

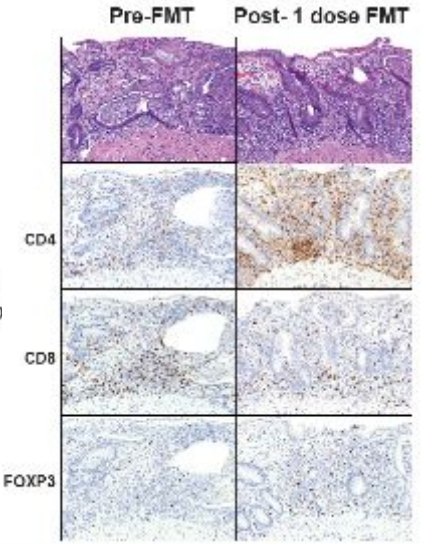
We already have evidence that use of fecal microbiome transplant (FMT) could be helpful in treating immunotherapy toxicity

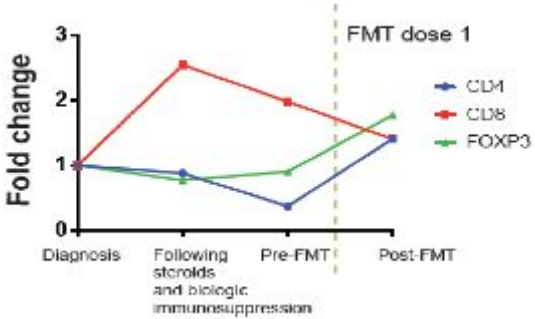


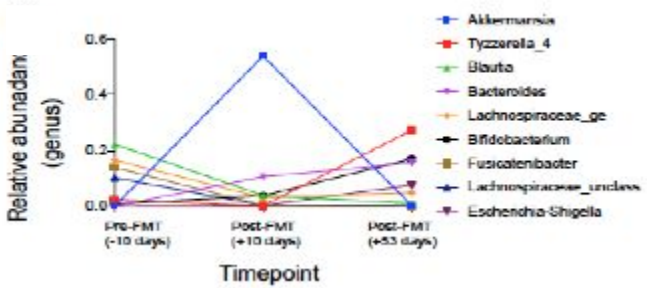
50 yo female with metastatic urothelial cancer was treated with aCTLA-4 + a PD-1 and developed colitis refractory to steroids and aTNF

She was treated with FMT from a healthy donor and had complete resolution of all symptoms









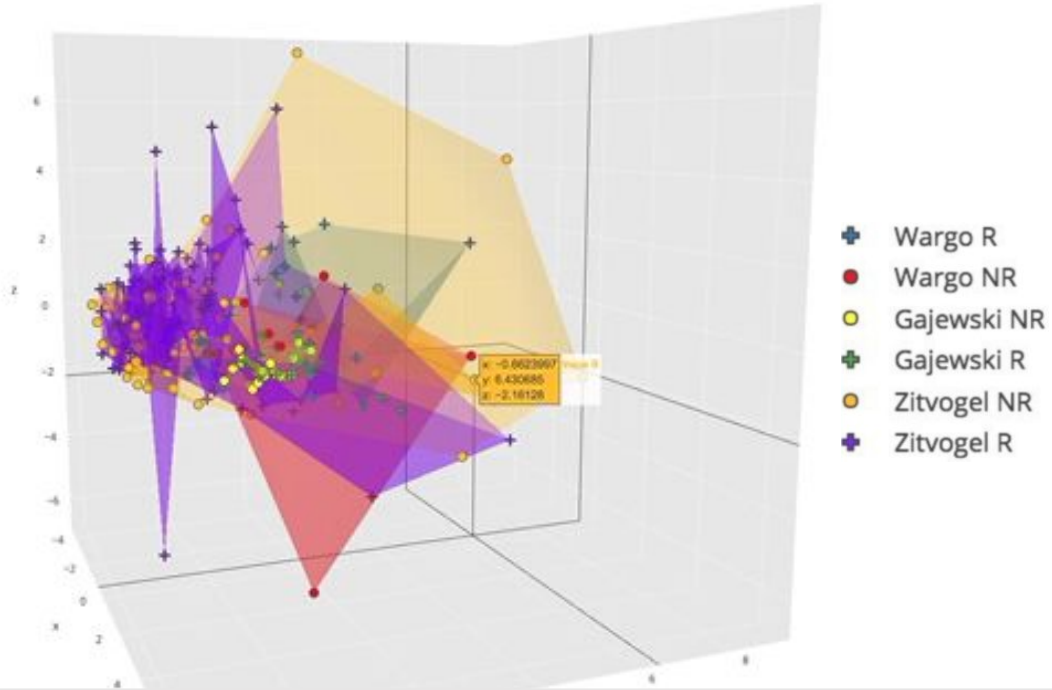
Mimi Wang MD PhD

*Wang et al, manuscript submitted * PLEASE **DO NOT POST** **

Rob Jenq MD

Can we engineer optimal microbial consortia to enhance responses to immunotherapy?

Though giving microbial consortia is the ultimate goal, complexities exist as optimal formulation for consortia is unknown (# of taxa, which ones, etc)



There is modest overlap between taxa associated with response to ICB in each of the published cohorts

- *Related in part to differences in sequencing, potential geographic differences*
- *Function may be more important than phylogeny*

Results from FMT and other

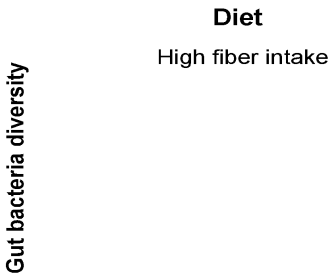
However some data will be presented at SITC on the use of microbial consortia from Kenya Honda's group "A rationally-designed consortium of human gut commensals induces CD8 T cells and modulates host and anti-cancer immunity" – Poster session (P574)

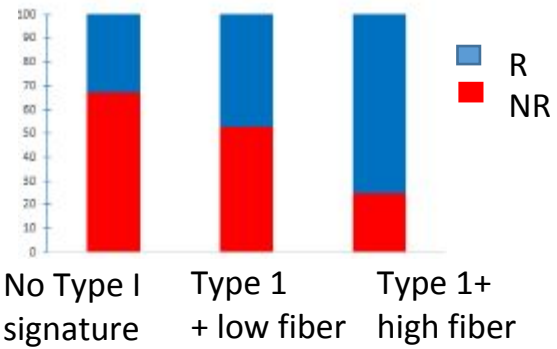
Matson et al, Routy et al, Gopalakrishnan et al Science 2018; integrated analysis courtesy of Vastbiome * PLEASE **DO NOT POST** *

Can we devise rational dietary strategies to enhance responses to immunotherapy?

We have provocative data in a human cohort demonstrating that patients with a high fiber diet have better responses to checkpoint blockade

Patients with a high fiber diet had higher diversity in the gut microbiome (with higher abundance of "favorable" gut bacteria)



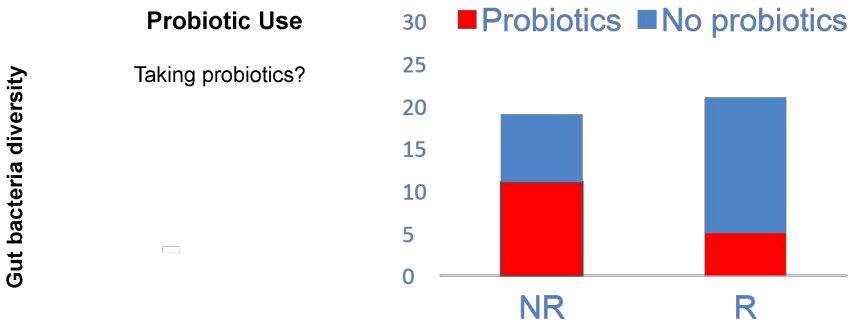


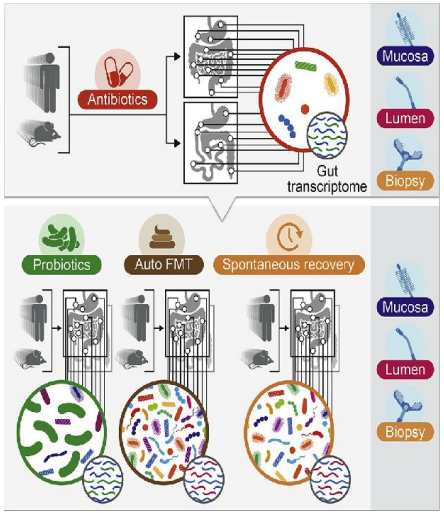
Importantly, parallel data exists in pre-clinical models suggesting that modulating fiber intake may enhance responses to immune checkpoint blockade (Vetizou, Trinchieri et al)

We are working together to better understand this – and will also be running dietary intervention trials to assess impact on microbiota, immunity, and response to immunotherapy

Importantly, we and others are also studying the impact of other factors (such as stress, medications, and probiotic use) on the microbiome and response

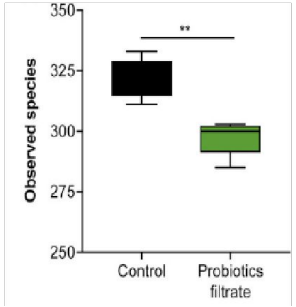
In our cohort, 42% of our patients reported taking probiotics, and this was associated with a LOWER diversity in the gut microbiome and a lower likelihood of response to anti-PD-1 therapy





Post-antibiotic gut mucosal microbiome reconstitution is impaired by probiotics

Suez....Segal, Elinav *Cell* 2018



This is consistent with recently published data in Cell suggesting that probiotics hinder recovery of gut flora after antibiotic use

Carrie Daniel MacDougall PhD
Jen McQuade MD

*Spencer et al, confidential unpublished data * PLEASE DO NOT POST**

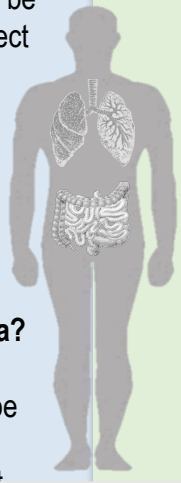
Chris Spencer PhD
Deepak Gopalakrishnan PhD

Prior to treatment

Patients
- What patient population to treat? Treatment naïve or refractory?
- Should the microbiome be profiled to stratify / select patients?

Pre-conditioning regimen
- Do we need to pre-treat the gut with antibiotics to facilitate engraftment?

How should we optimally modulate the gut microbiota?
- FMT?
- How should FMT be administered?
- How do we select
- Should patient fecal material be "banked" for later auto-FMT?
- Diet?
- Designer Consortia?
- Phage / antibiotics / other?



During therapy

What therapy should we combine with modulation of the gut microbiome?
- Immune checkpoint blockade (anti-PD-1)?
- Other forms of immunotherapy?
- Other therapy?

How do we optimally monitor patients during therapy?
- Microbiome analyses to assess engrafment / function?
- Immune profiling?
- Peripheral blood
- Tumor

engraftment?
- Should we recommend dietary changes?
- Any medications to avoid?

Assessing impact

What are appropriate primary endpoints for such studies?
- Safety and tolerability
- Engraftment
- Others?

What are appropriate secondary endpoints?
- Response
- Radiographic (RECIST and / or irRC)
- Rate of complete responses

neoadjuvant therapy)
- Toxicity
- Novel markers (ctDNA, immunophenotyping)

Long-term effects

Durability of engraftment
- How durable is engraftment?
- What microbes / functional phenotypes in gut microbiota are associated with responses? And can these be used to design consortia?

Overall responses
- What is impact on overall and disease-specific survival?

Toxicity
- Can we uncouple toxicity and response to immunotherapy?

s with
FMT?
- Obesity?
- Depression?
- Any potentially favorable traits?

There are a lot of considerations as we move forward with these approaches!

Conclusions and potential implications of these findings:

- There is increasing evidence for the role of the microbiome in health and disease (in the gut and other sites), with evidence that microbiota may influence immunity and responses to cancer therapy
- Microbiota within tumors and/or the gut of patients may serve as a biomarker of response to cancer therapy, though this needs to be validated in larger cohorts (with standardized approaches to characterize the microbiome)
- Efforts to modulate microbes to enhance response to cancer are currently underway, though optimal means to do this remain incompletely understood
- Development of optimal strategies will rely on a deep mechanistic understanding of how the microbiome influences therapeutic responses – as well as an appreciation of all of the factors that influence the microbiota (including diet, medications, and other factors) – *with critical insights gained through collaboration*

Cogdill, Andrews, Wargo *British Journal of Cancer (submitted)*

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- **Beth Helmink MD PhD**
- **Alexandria P. Cogdill MS (PhD candidate)**
- Robert Szczepaniak-Sloane BS (PhD candidate)
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- **Sangeetha Reddy MD PhD**
- **Liz Burton MBA**

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- **Ravid Straussman MD PhD, Yardena Samuels PhD**
- **Parker Institute Investigators**

MDACC Collaborators

- **Jim Allison PhD, Pam Sharma MD PhD**
- **Michael Davies MD PhD, Jeff Gershenwald MD**
- **Patrick Hwu MD, other Melanoma Med Onc Facutly / Staff**
- Jeff Lee MD, Merrick Ross MD, other Surg Onc Faculty / Staff
- Andrew Futreal PhD, **Carrie Daniel-MacDougall PhD**
- Michael Tetzlaff MD PhD, Alex Lazar MD, **Jen McQuade MD MS**
- Robert Jenq MD PhD, other MDACC faculty / staff

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Baylor CMMR

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