Microbiome and Immune Tolerance

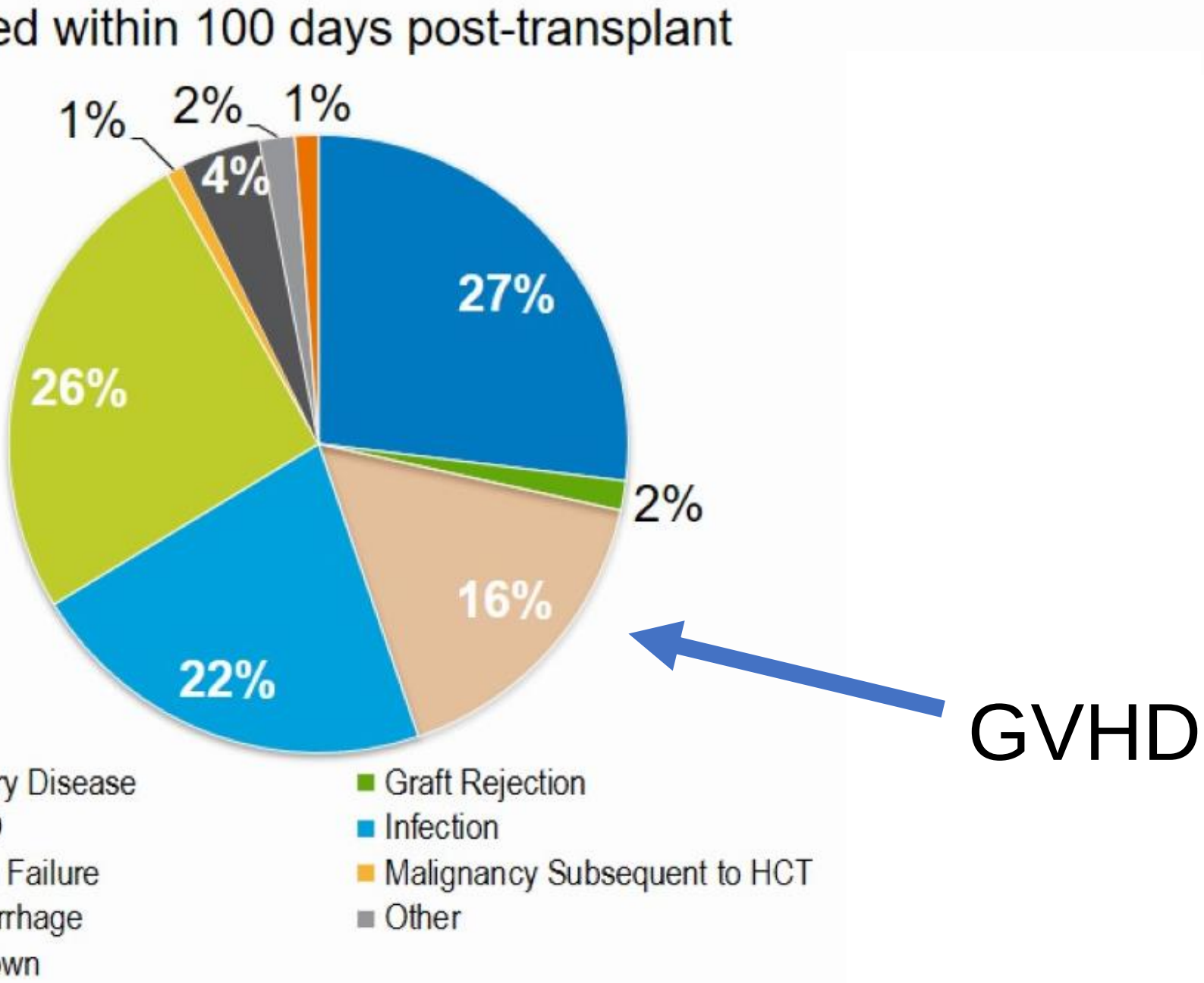
If we can't live without it, how best to live with it?

Lessons learned from allogeneic hematopoietic cell transplantation

Robert Jenq, MD

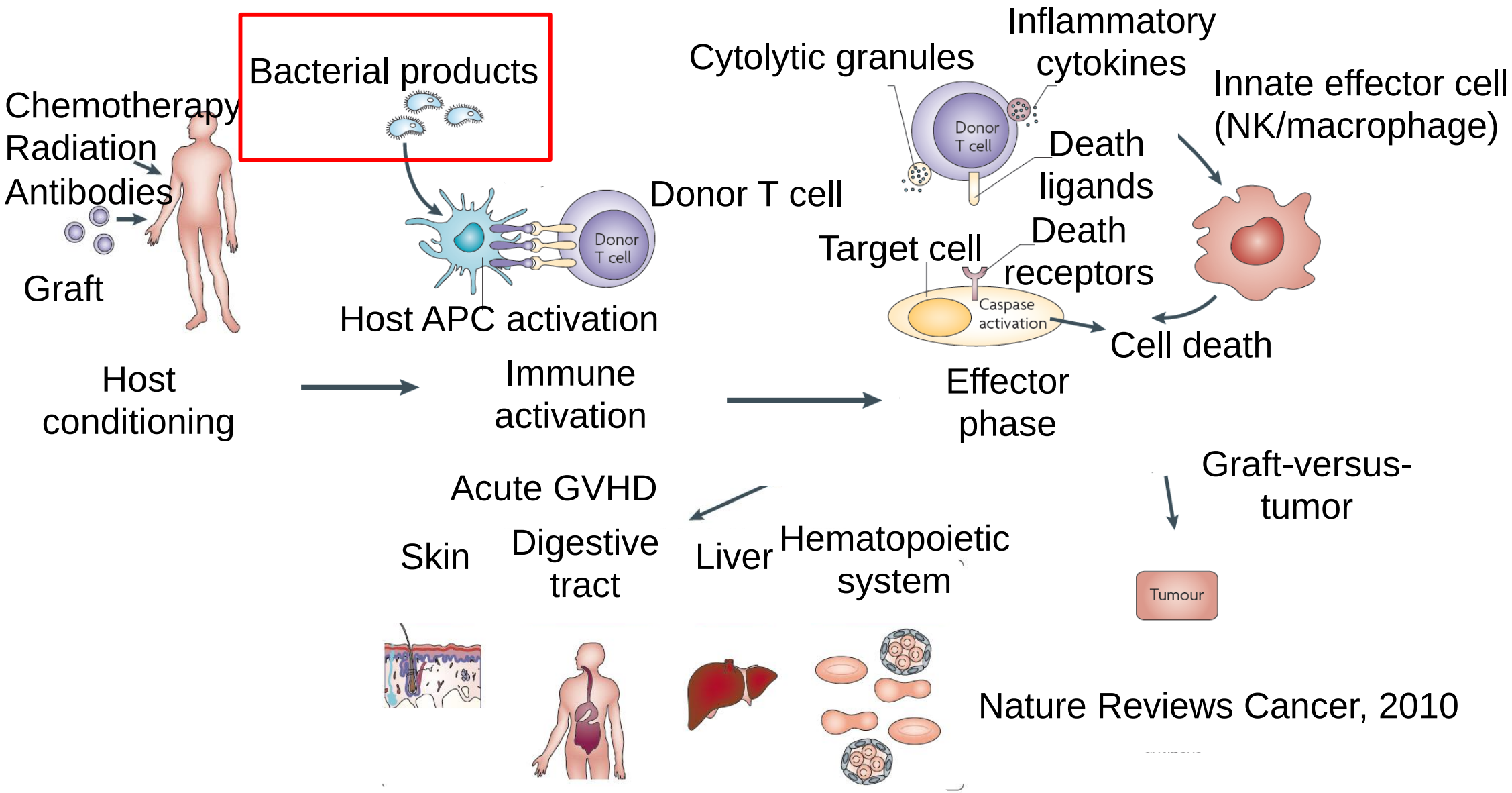
THE UNIVERSITY OF TEXAS
MD Anderson Cancer Center
Making Cancer History®

Causes of Death after Adult (age ≥18) Unrelated Donor HCT in the US, 2017-2018

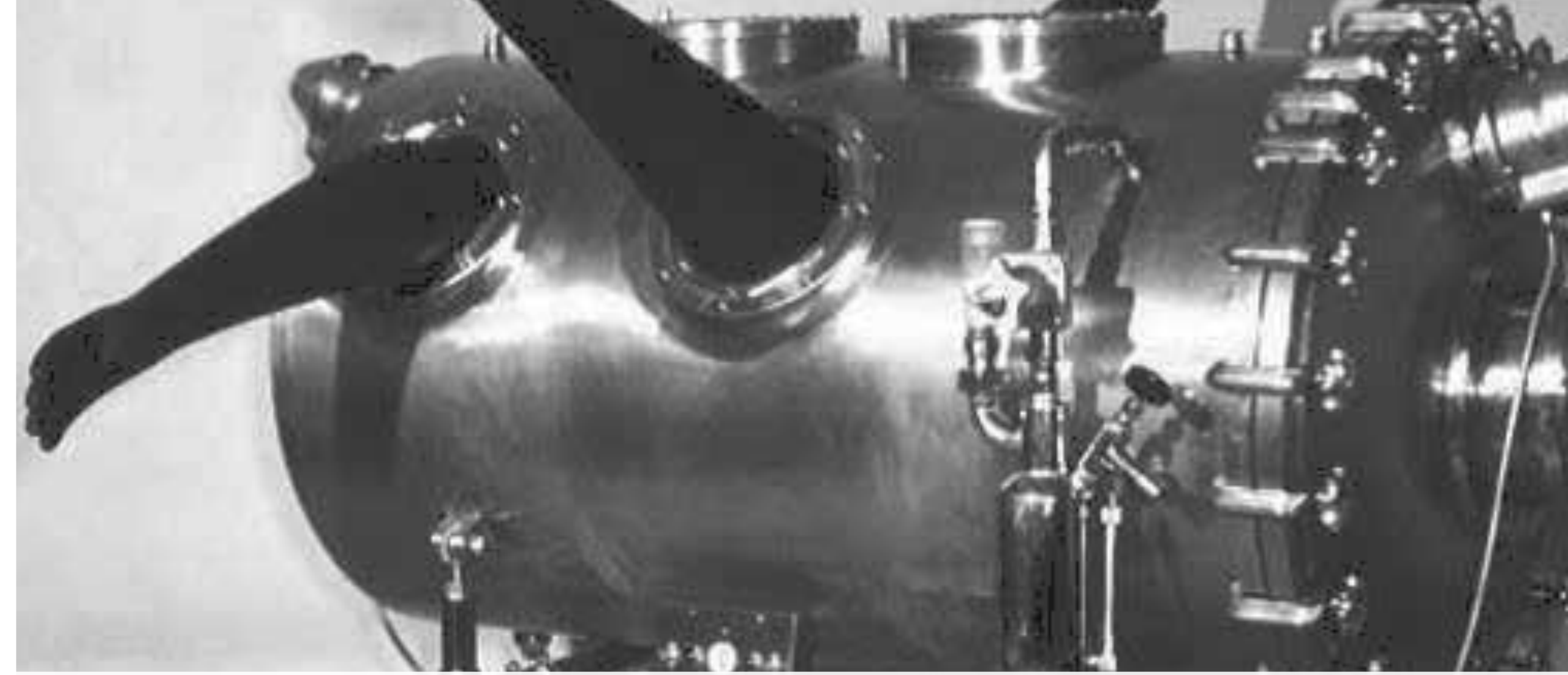


CIBMTR
CENTER FOR INTERNATIONAL BLOOD & MARROW TRANSPLANT RESEARCH

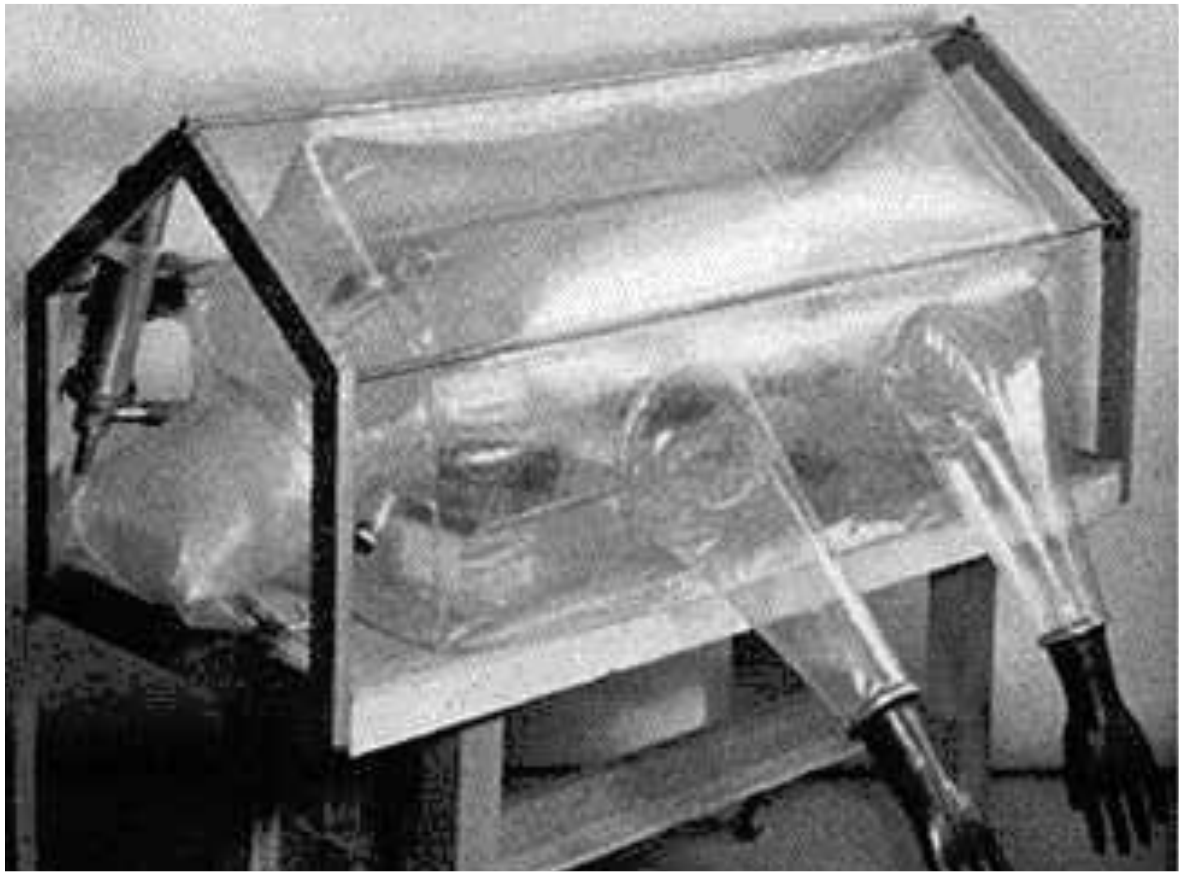
BMT scientists and physicians – microbiome pioneers



Nature Reviews Cancer, 2010



Steam sterilizable metal isolator (Notre Dame University, 1928)



1962 Trexler isolator

2021 Class Biologically Clean (CBC) flexible film isolators

www.cleanairandcontainment.com

flexiblefilmisolators.com

Bacteria contribute to the pathophysiology of GVHD

Mortality and Gross Pathology of Secondary Disease in Germfree Mouse Radiation Chimeras[1]

J. MIRIAM JONES[2], RAPHAEL WILSON, AND PATRICIA M. BEALMEAR

1971

Mitigation of Secondary Disease of Allogeneic Mouse Radiation Chimeras by Modification of the Intestinal Microflora [1]

D. W. van Bekkum, J. Roodenburg, P. J. Heidt, *and* D. van der Waaij [2]

1974

Fred Hutch transplantation clinic in the 1980s

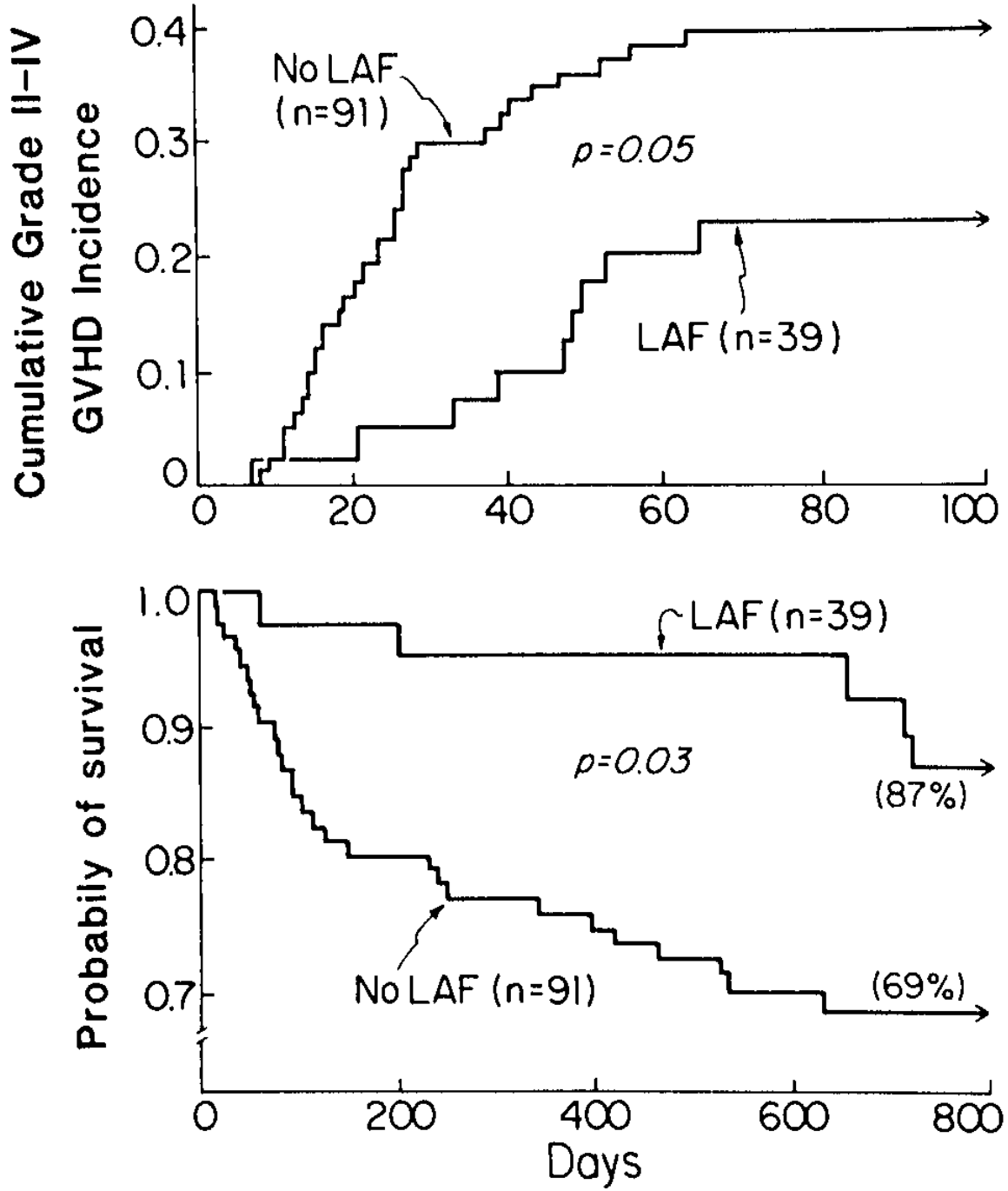
www.fredhutch.org/

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

RAINER STORB, M.D., ROSS L. PRENTICE, PH.D., C. DEAN BUCKNER, M.D., R. A. CLIFT, F.I.M.L.S., FRED APPELBAUM, M.D., JOACHIM DEEG, M.D., KRISTINE DONEY, M.D., JOHN A. HANSEN, M.D., MARK MASON, JEAN E. SANDERS, M.D., JACK SINGER, M.D., KEITH M. SULLIVAN, M.D., ROBERT P. WITHERSPOON, M.D., AND E. DONNALL THOMAS, M.D.

- 130 patients with severe aplastic anemia
- Conditioned with cyclophosphamide followed by marrow from HLA-identical siblings
- 39 randomly assigned to a protective environment:
  - Laminar airflow
  - “Sterile" food
  - Oral nonabsorbable antibiotics and skin cleansing
  - Minimum of 50 days


Cumulative Grade II–IV
GVHD Incidence
0.4
0.3
0.2
0.1
0
No LAF
(n=91)
p=0.05
LAF (n=39)
0
20
40
60
80
100
Probabily of survival
1.0
0.9
0.8
0.7
LAF (n=39)
p=0.03
(87%)
No LAF (n=91)
(69%)
0
200
400
600
800
Days

Controversies in intestinal bacterial decontamination in allo-HCT

- Clinical studies initially suggested a benefit from near-total bacterial decontamination (Storb et al., 1983)
- Later studies, however, showed no clear benefit (Peterson et al., 1987; Passweg et al., 1998; Russell et al., 2000).

The NEW ENGLAND JOURNAL of MEDICINE

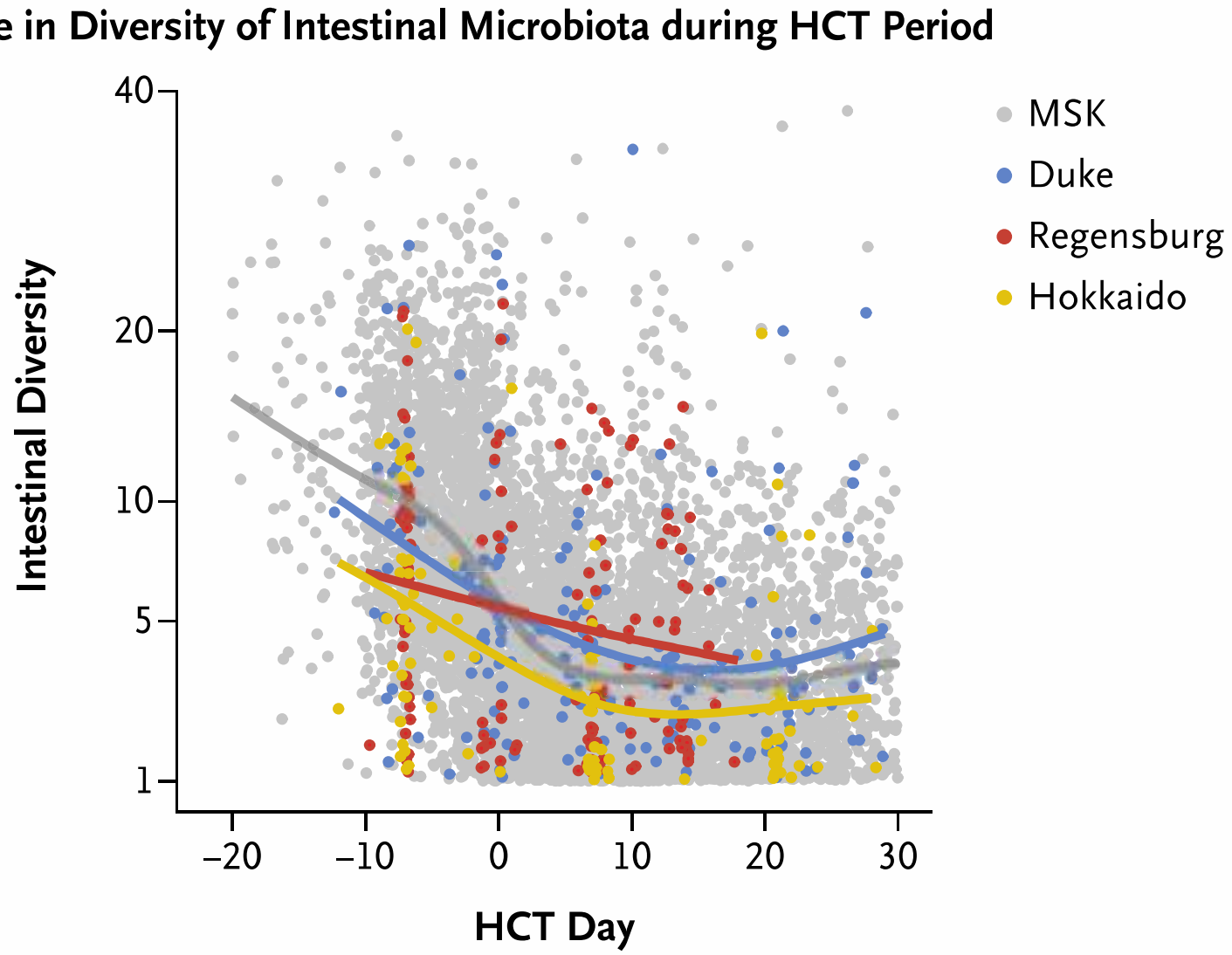
ORIGINAL ARTICLE

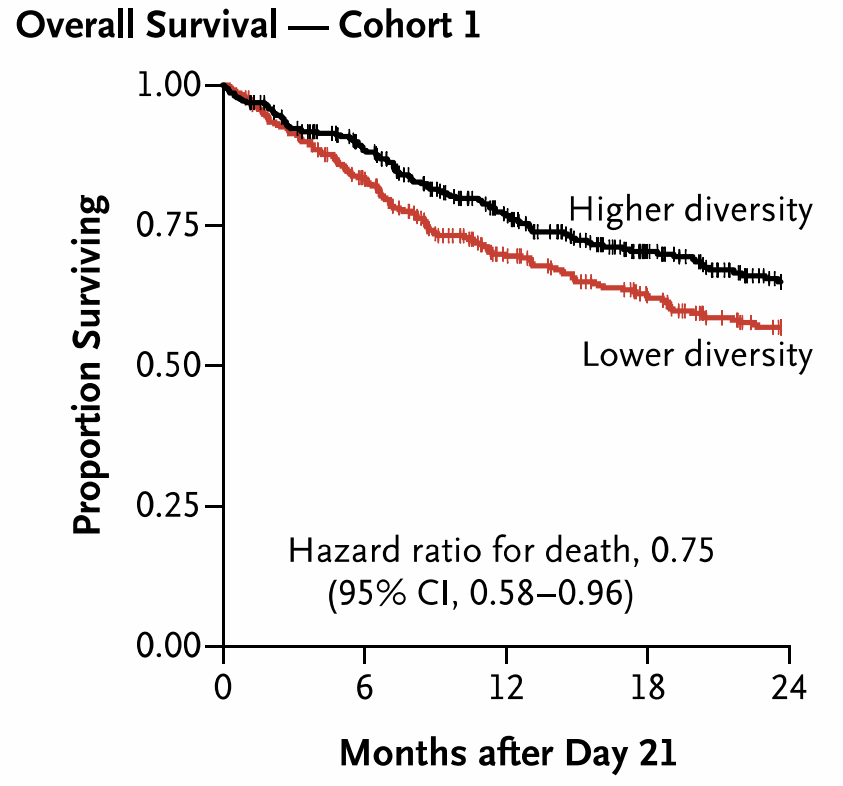
Microbiota as Predictor of Mortality in Allogeneic Hematopoietic-Cell Transplantation

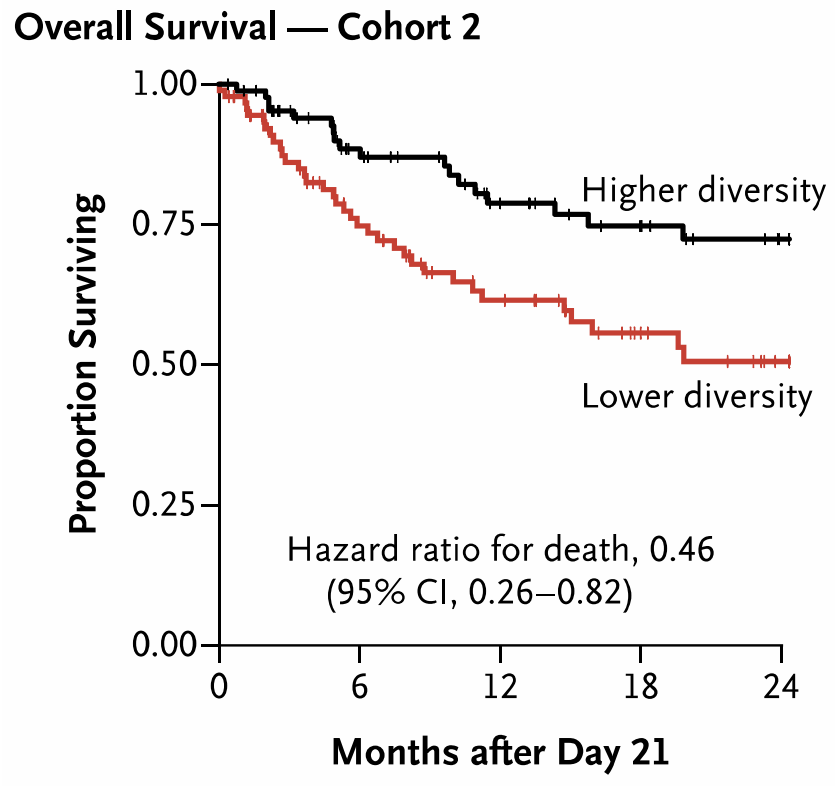
J.U. Peled, A.L.C. Gomes, S.M. Devlin, E.R. Littmann, Y. Taur, A.D. Sung, D. Weber, D. Hashimoto, A.E. Slingerland, J.B. Slingerland, M. Maloy, A.G. Clurman, C.K. Stein-Thoeringer, K.A. Markey, M.D. Docampo, M. Burgos da Silva, N. Khan, A. Gessner, J.A. Messina, K. Romero, M.V. Lew, A. Bush, L. Bohannon, D.G. Brereton, E. Fontana, L.A. Amoretti, R.J. Wright, G.K. Armijo, Y. Shono, M. Sanchez-Escamilla, N. Castillo Flores, A. Alarcon Tomas, R.J. Lin, L. Yáñez San Segundo, G.L. Shah, C. Cho, M. Scordo, I. Politikos, K. Hayasaka, Y. Hasegawa, B. Gyurkocza, D.M. Ponce, J.N. Barker, M.-A. Perales, S.A. Giralt, R.R. Jenq, T. Teshima, N.J. Chao, E. Holler, J.B. Xavier, E.G. Pamer, and M.R.M. van den Brink

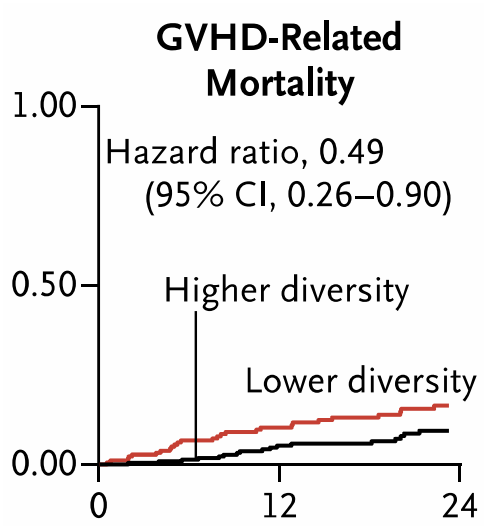
RESULTS

We profiled 8767 fecal samples obtained from 1362 patients undergoing allogeneic hematopoietic-cell transplantation at the four centers. We observed patterns of micro-




Overall Survival — Cohort 1
Proportion Surviving
1.00
0.75
0.50
0.25
0.00
Higher diversity
Lower diversity
Hazard ratio for death, 0.75
(95% CI, 0.58–0.96)
0
6
12
18
24
Months after Day 21

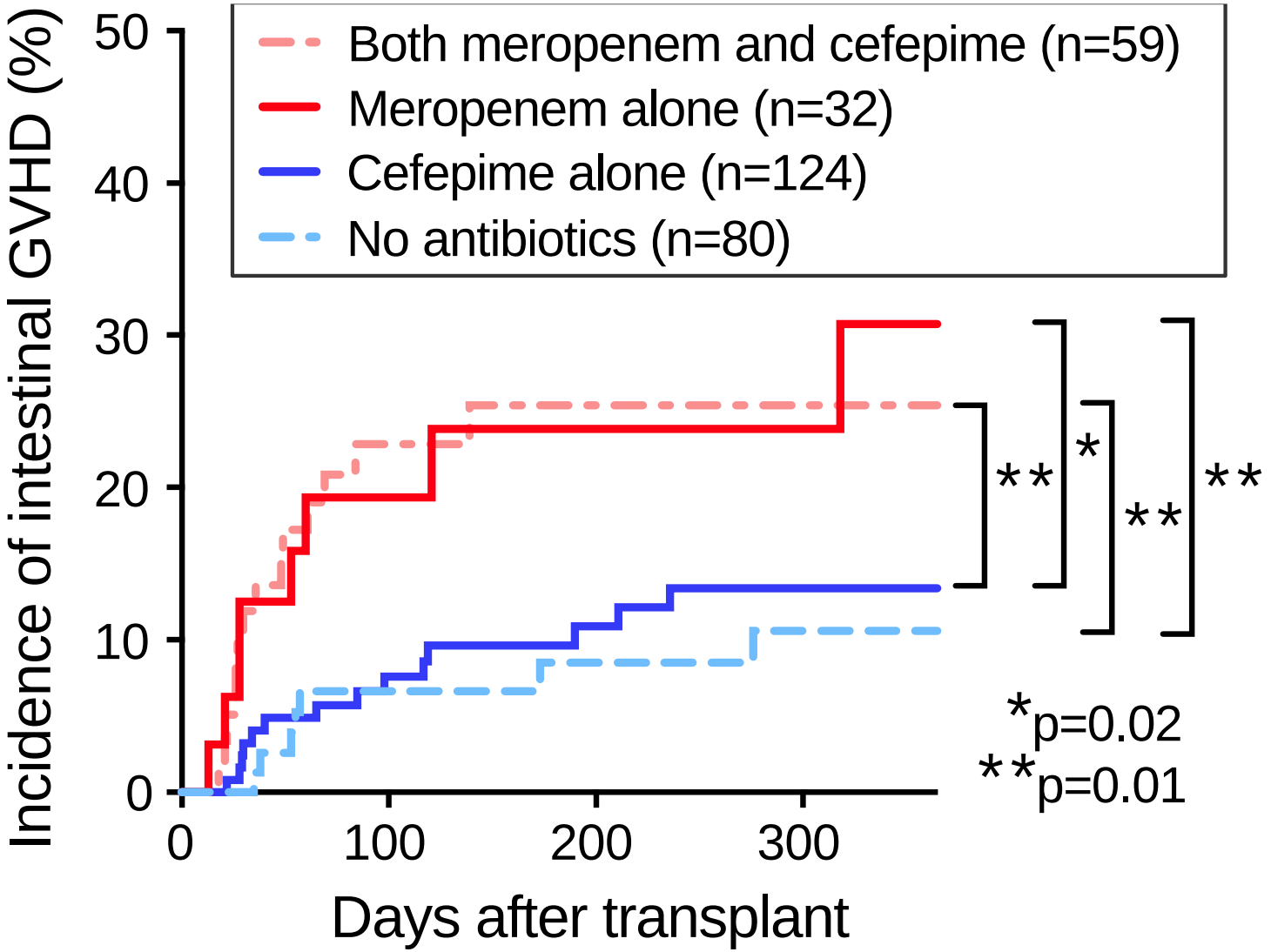

Overall Survival — Cohort 2
Proportion Surviving
1.00
0.75
0.50
0.25
0.00
Higher diversity
Lower diversity
Hazard ratio for death, 0.46
(95% CI, 0.26–0.82)
0
6
12
18
24
Months after Day 21


GVHD-Related Mortality
1.00
0.50
0.00
Hazard ratio, 0.49
(95% CI, 0.26–0.90)
Higher diversity
Lower diversity
0
12
24

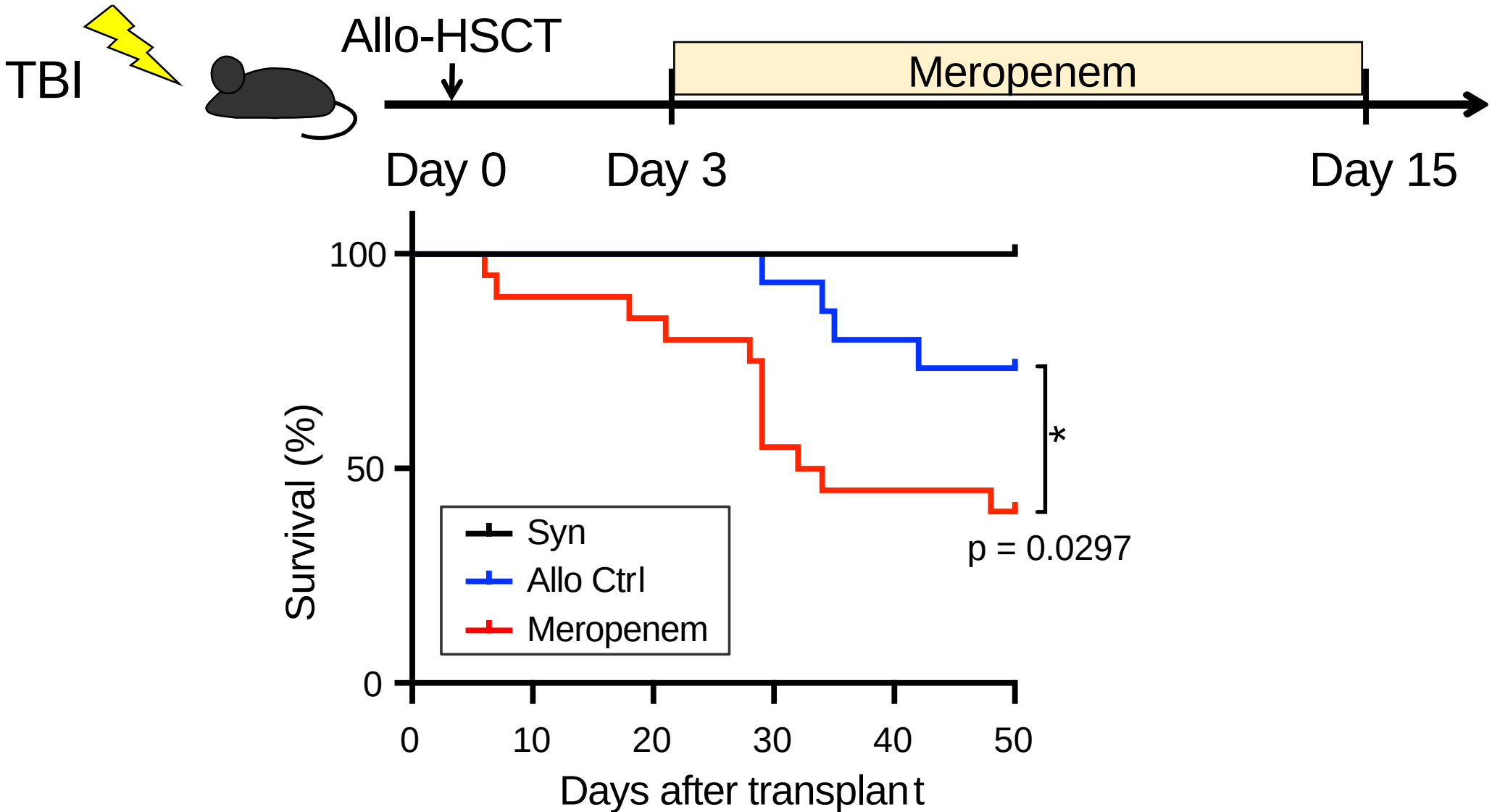
	MSKCC Estimate ± S.E.	Regensburg Estimate ± S.E.	Duke Estimate ± S.E.	Hokkaido Estimate ± S.E.
Δt	**-0.33 ± .02**	-0.10 ± 0.17	**-0.15 ± 0.06**	-0.01 ± 0.06
cefepime	0.07 ± .07	-	-0.13 ± 0.22	0.18 ± 0.19
doripenem	-	-	-	0.11 ± 0.23
meropenem	0.08 ± 0.21	-0.02 ± 0.24	-0.51 ± 0.33	**-0.36 ± 0.15**
piperacillin-tazobactam	**-0.13 ± 0.05**	-0.24 ± se 0.29	-0.35 ± 0.28	-
teicoplanin	-	-	-	-0.15 ± 0.12

	MSKCC Estimate ± S.E.	Regensburg Estimate ± S.E.	Duke Estimate ± S.E.	Hokkaido Estimate ± S.E.
Δt	**-0.33 ± .02**	-0.10 ± 0.17	**-0.15 ± 0.06**	-0.01 ± 0.06
cefepime	0.07 ± .07	-	-0.13 ± 0.22	0.18 ± 0.19
doripenem	-	-	-	0.11 ± 0.23
meropenem	0.08 ± 0.21	-0.02 ± 0.24	-0.51 ± 0.33	**-0.36 ± 0.15**
piperacillin-tazobactam	**-0.13 ± 0.05**	-0.24 ± se 0.29	-0.35 ± 0.28	-
teicoplanin	-	-	-	-0.15 ± 0.12

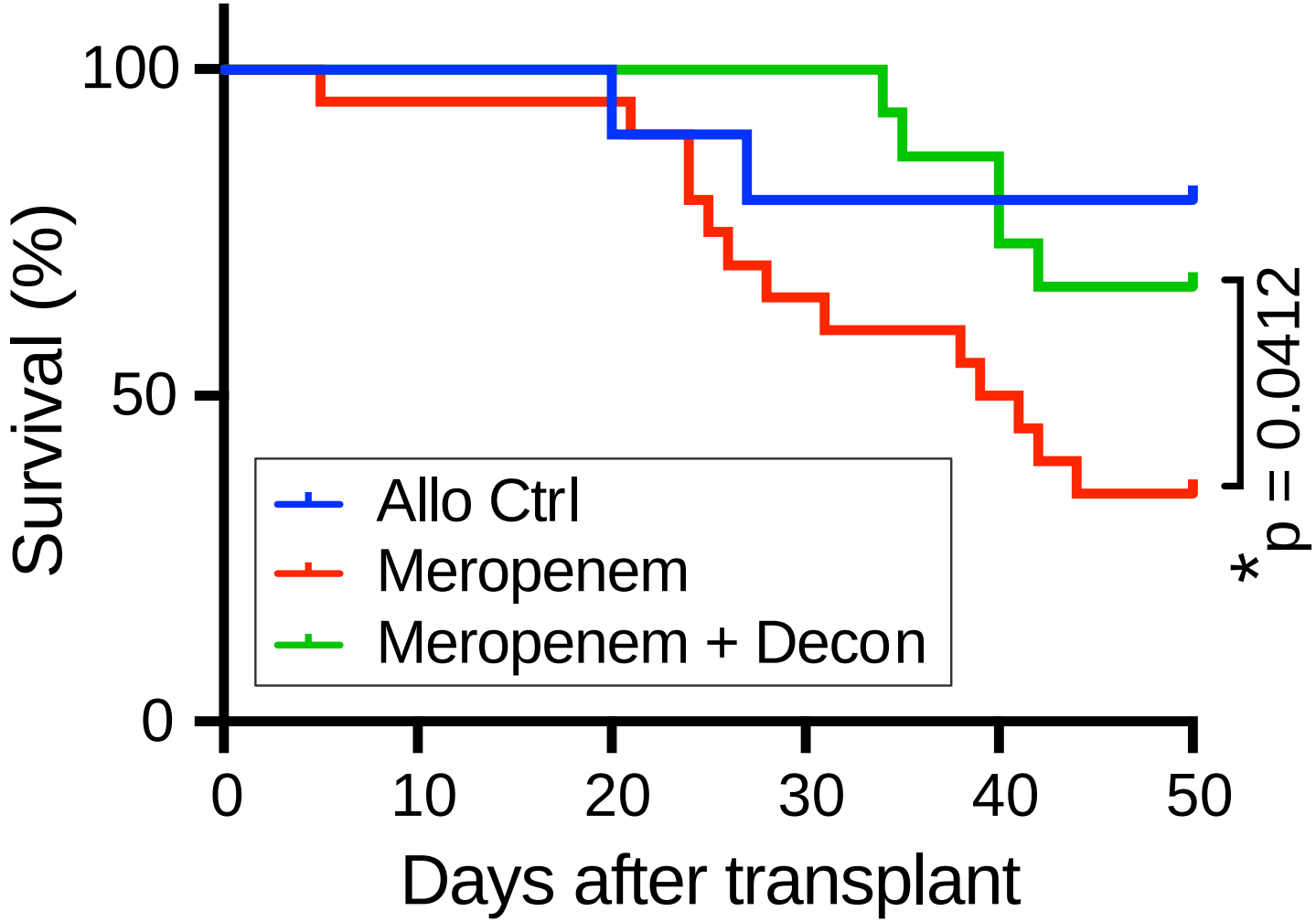
Meropenem treatment is associated with aggravated intestinal GVHD



Meropenem treatment is associated with aggravated intestinal GVHD



Decontamination can rescue meropenem-treated GVHD mice



Intestinal bacteria

Phylum	Class	Order	Genus	Gram stain	Type of anaerobe
Firmicutes	Bacilli	Bacillales	Gemella Staphylococcus	+	Facultative
		Lactobacillales	Enterococcus Lactobacillus Streptococcus		
	Clostridia	Clostridiales	Blautia Clostridium Eubacterium Faecalibacterium Ruminococcus	+	Obligate
	Erysipelotrichia	Erysipelotrichiales	Erysipelatoclostridium Holdemania	+	Obligate
	Negativicutes	Selenomonadales	Acidaminococcus Megasphaera Phascolarctobacterium Veillonella	−	Obligate
Bacteroidetes	Bacteroidia	Bacteroidales	Bacteroides Prevotella	−	Obligate
Proteobacteria	Gamma-proteobacteria	Enterobacteriales	Escherichia Klebsiella	−	Facultative
Actinobacteria	Actinobacteria	Actinomycetales	Actinomyces	+	Facultative
		Bifidobacteriales	Bifidobacterium		
Verrucomicrobia	Verrucomicrobiae	Verrucomicrobiales	Akkermansia	−	Obligate

Intestinal bacteria

Phylum	Class	Order	Genus	Gram stain	Type of anaerobe
Firmicutes	Bacilli	Bacillales	Gemella Staphylococcus	+	Facultative
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	Erysipelotrichia	Erysipelotrichiales	Erysipelatoclostridium Holdemania	+	Obligate
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Bacteroidetes	Bacteroidia	Bacteroidales	Bacteroides Prevotella	−	Obligate
Proteobacteria	Gamma-proteobacteria	Enterobacteriales	Escherichia Klebsiella	−	Facultative
Actinobacteria	Actinobacteria	Actinomycetales	Actinomyces	+	Facultative
		Bifidobacteriales	Bifidobacterium		
Verrucomicrobia	Verrucomicrobiae	Verrucomicrobiales	Akkermansia	−	Obligate

Intestinal bacteria

Phylum	Class	Order	Genus	Gram stain	Type of anaerobe
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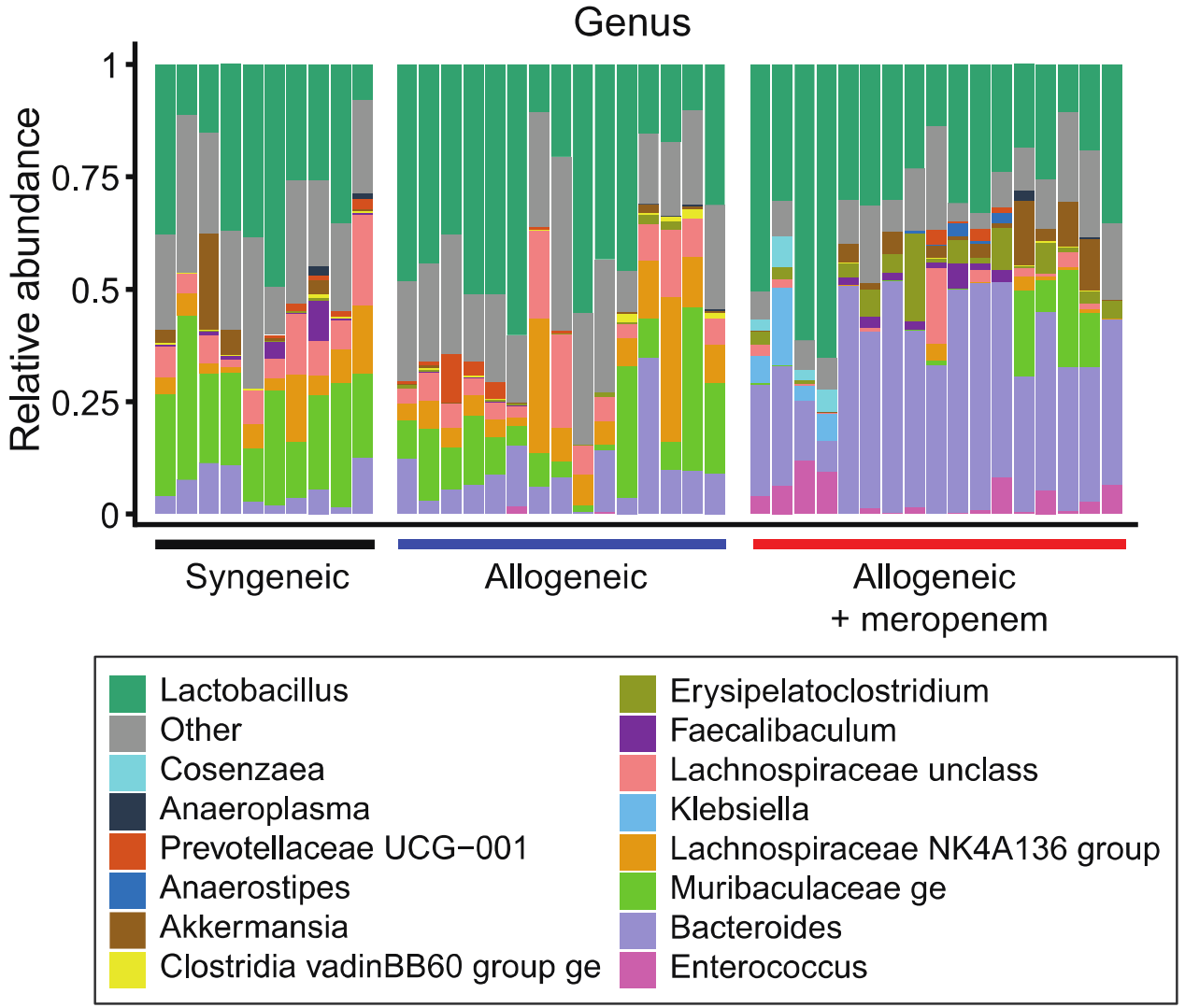
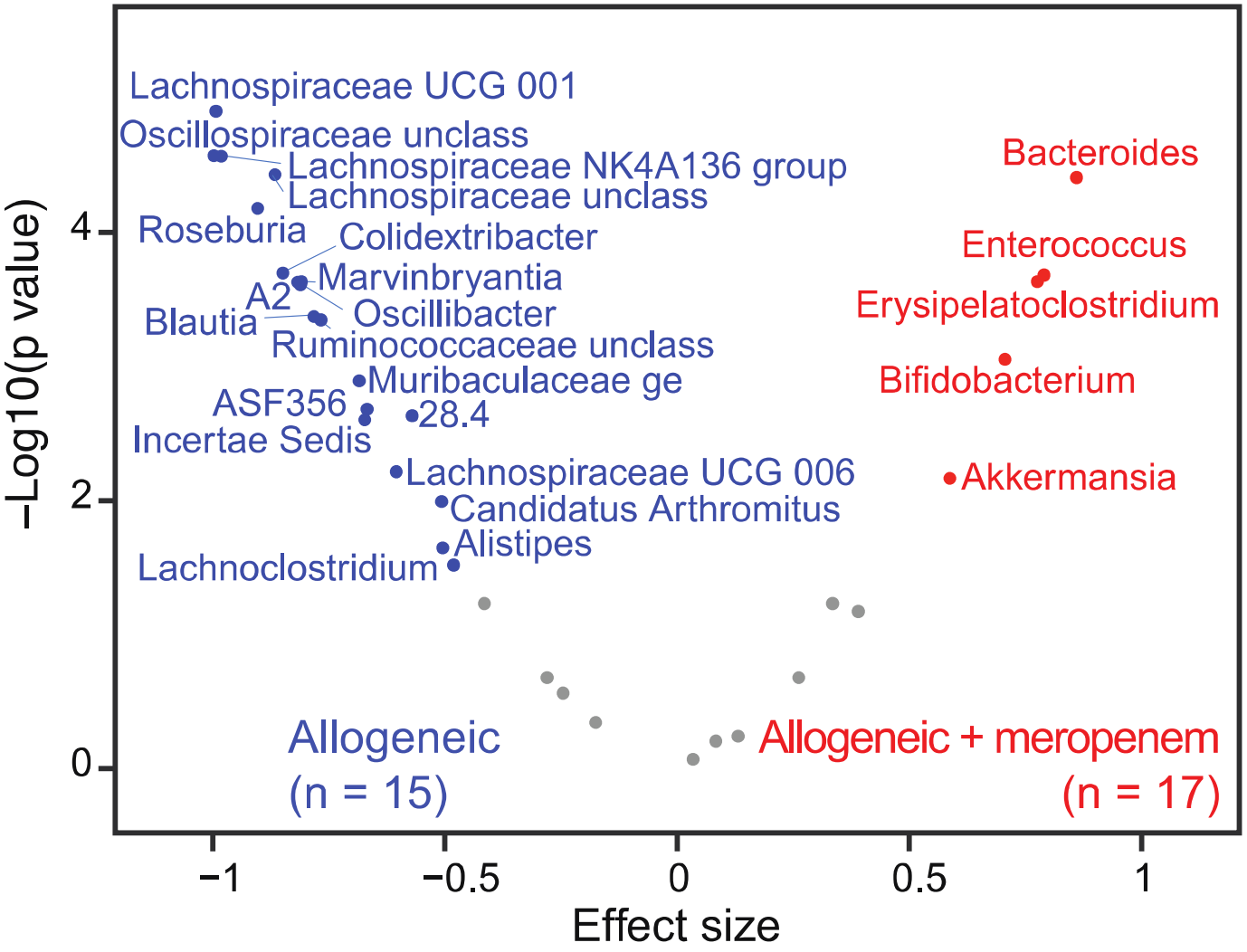
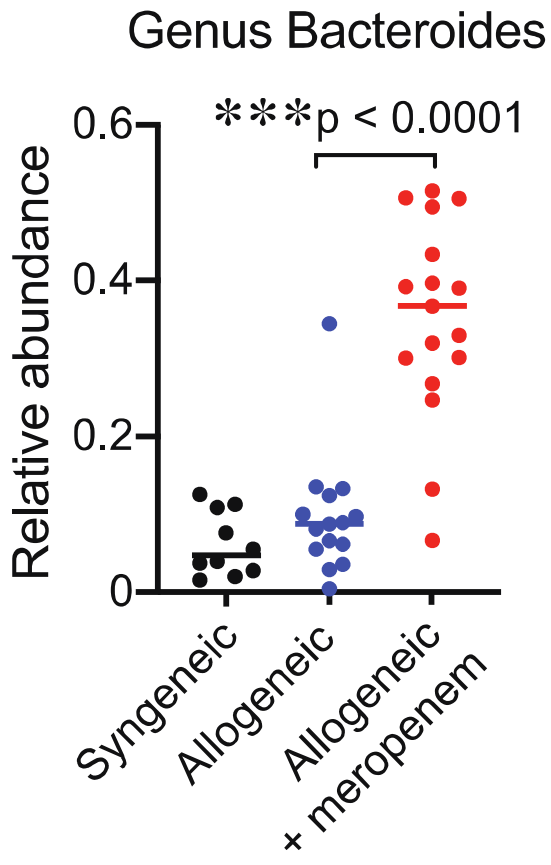
Bacteroidetes	Bacteroidia	Bacteroidales	Bacteroides Prevotella	−	Obligate
Proteobacteria	Gamma-proteobacteria	Enterobacteriales	Escherichia Klebsiella	−	Facultative
Actinobacteria	Actinobacteria	Actinomycetales	Actinomyces	+	Facultative
		Bifidobacteriales	Bifidobacterium		
Verrucomicrobia	Verrucomicrobiae	Verrucomicrobiales	Akkermansia	−	Obligate

B. thetaiotaomicron is increased in meropenem-treated GVHD mice

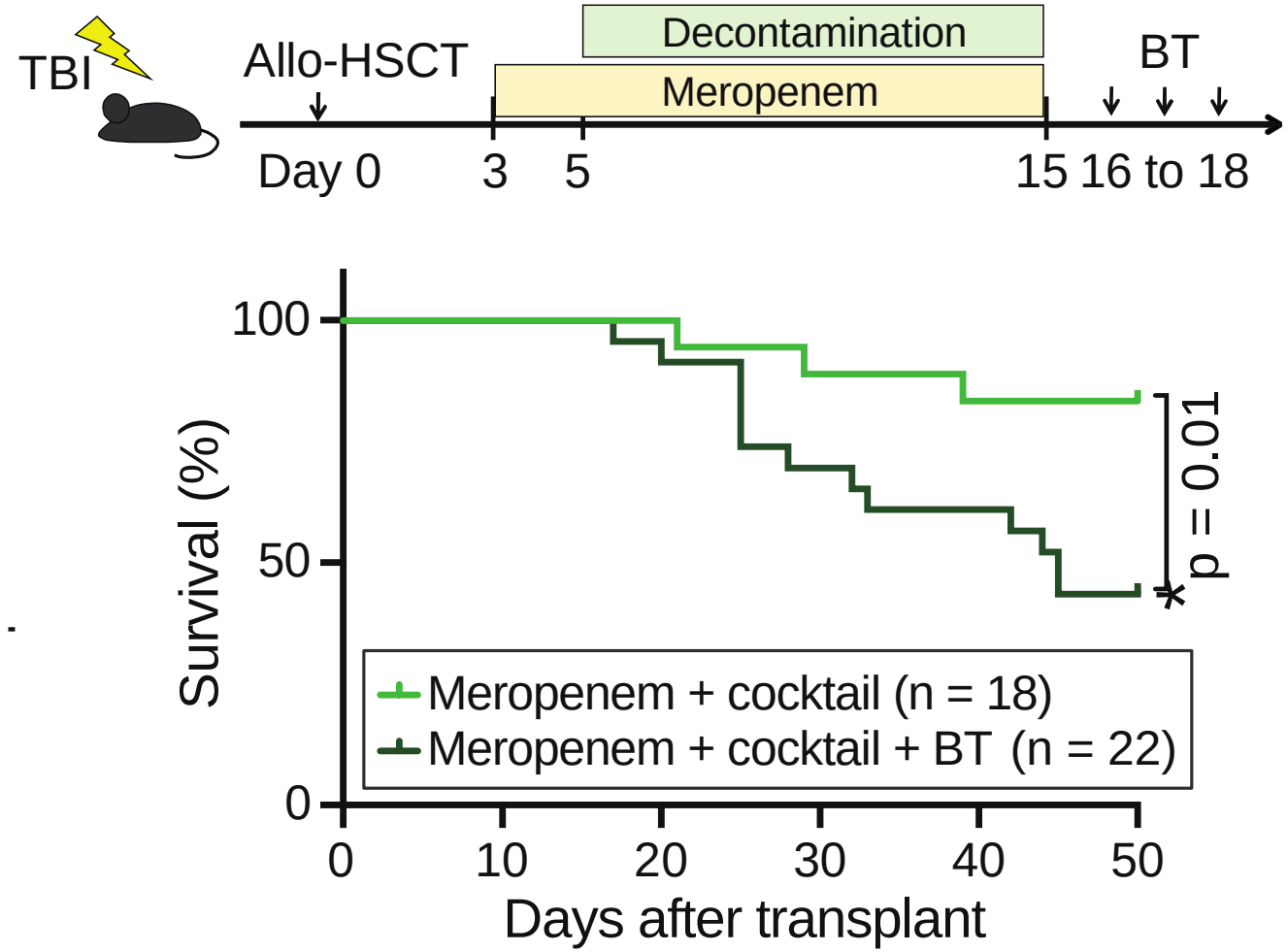


B. thetaiotaomicron is increased in meropenem-treated GVHD mice



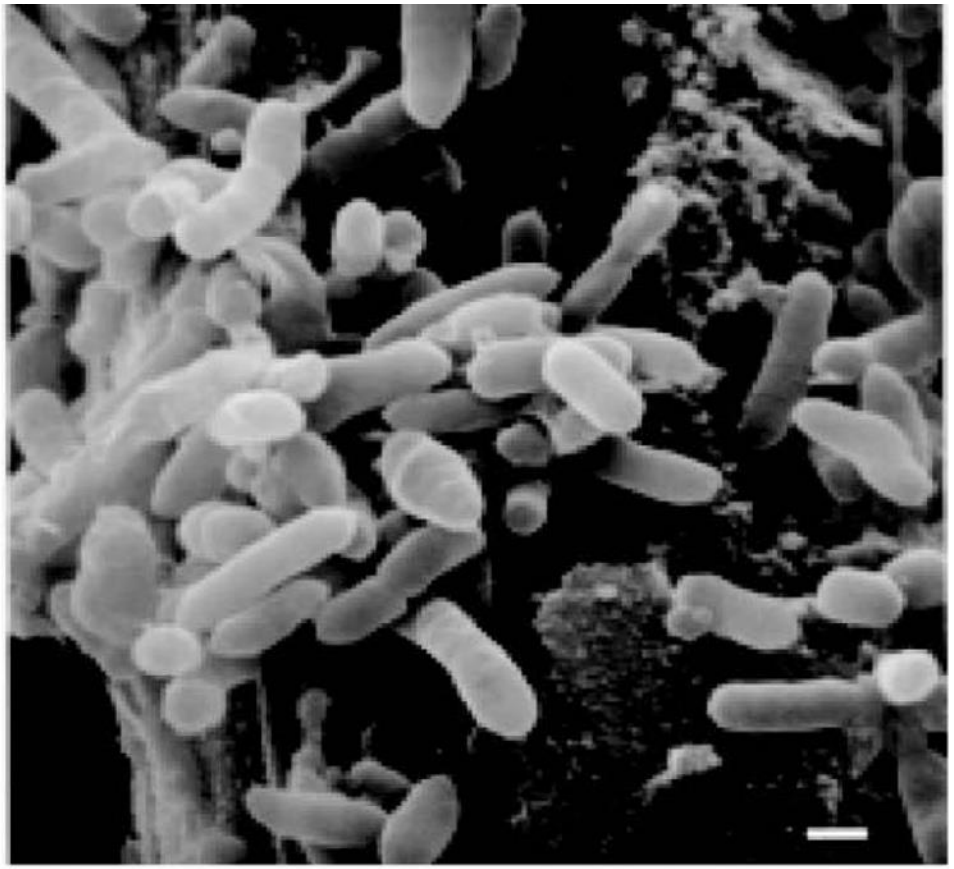


B. thetaiotaomicron aggravates GVHD when reintroduced to decontaminated mice



Bacteroides thetaiotaomicron

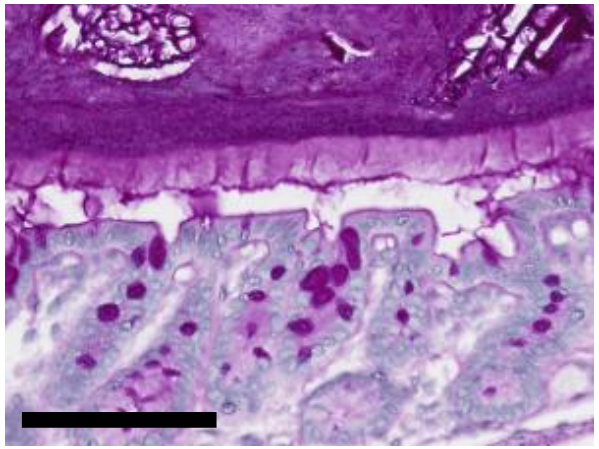
- A gram-negative obligate anaerobic bacteria
- Commensal bacteria in both mouse and human
- It has a broad ability to digest dietary fiber polysaccharides and glycans
- Ambient levels of metabolites, produced as a byproduct of substrate utilization, have been shown to play a major role in determining transcriptional profiles in *B. thetaiotaomicron*



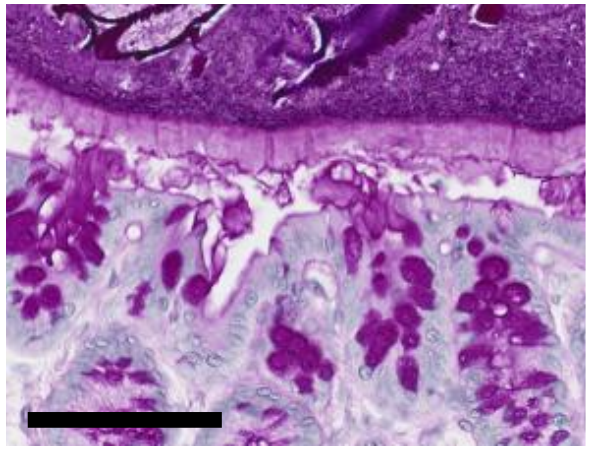
TerAvest MA et al; Biotechnol Bioeng, 2014

Meropenem arm showed thinned mucus layer and decontamination suppressed mucus degradation

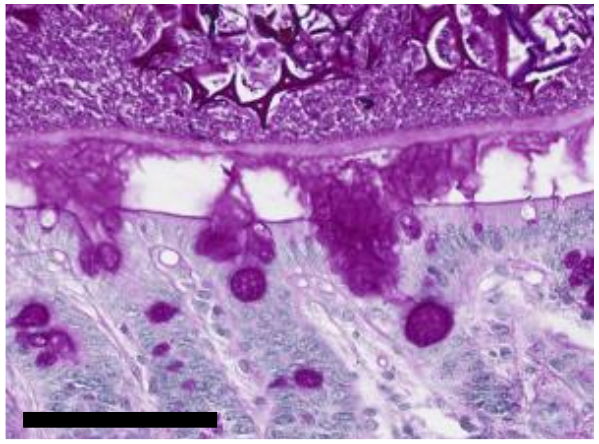
Syn



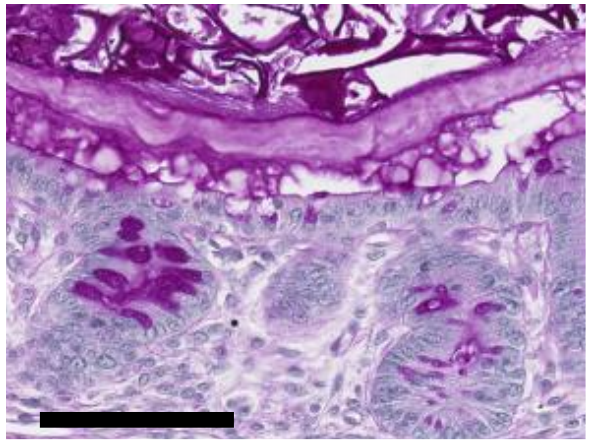
Allo Ctrl

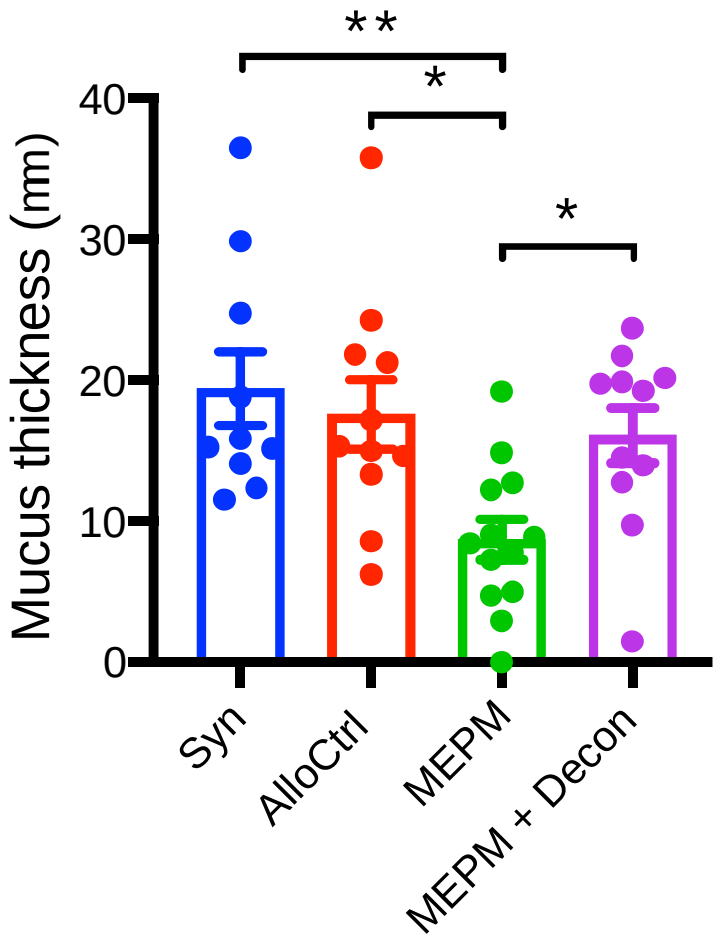


Meropenem



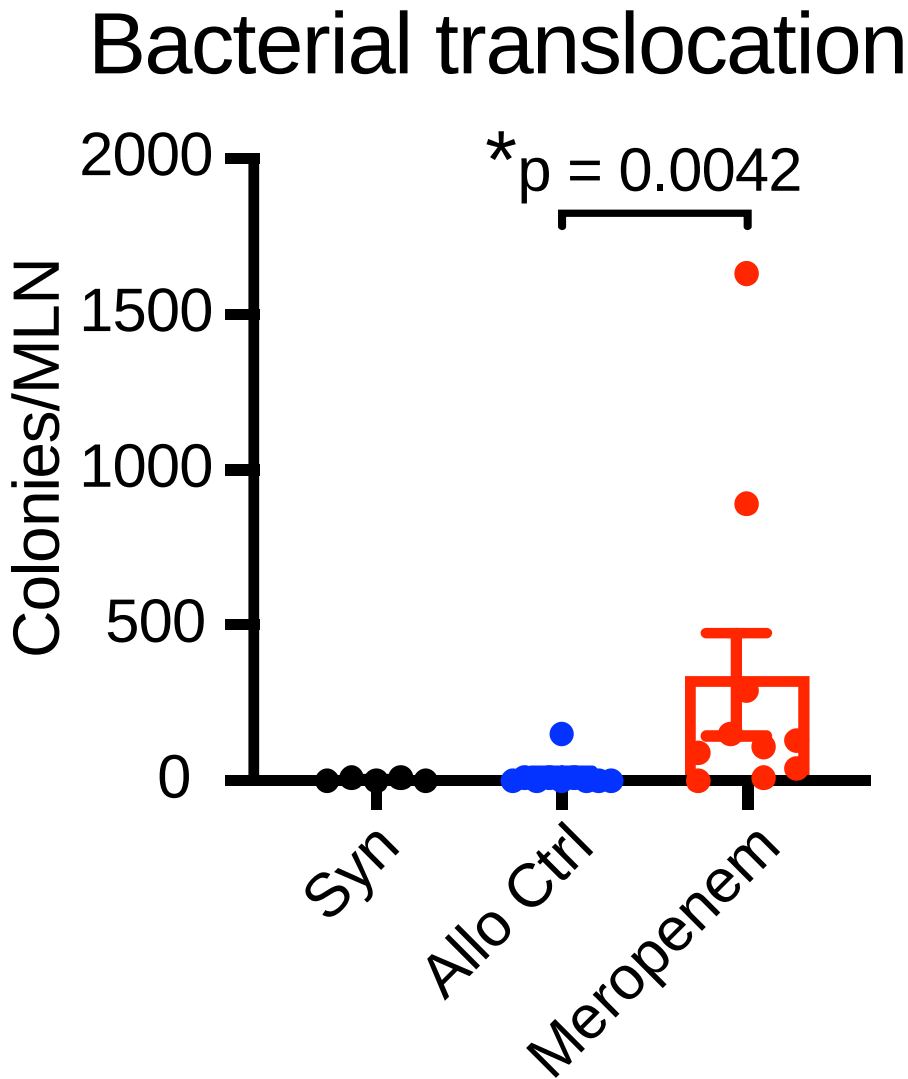
Meropenem + Decon



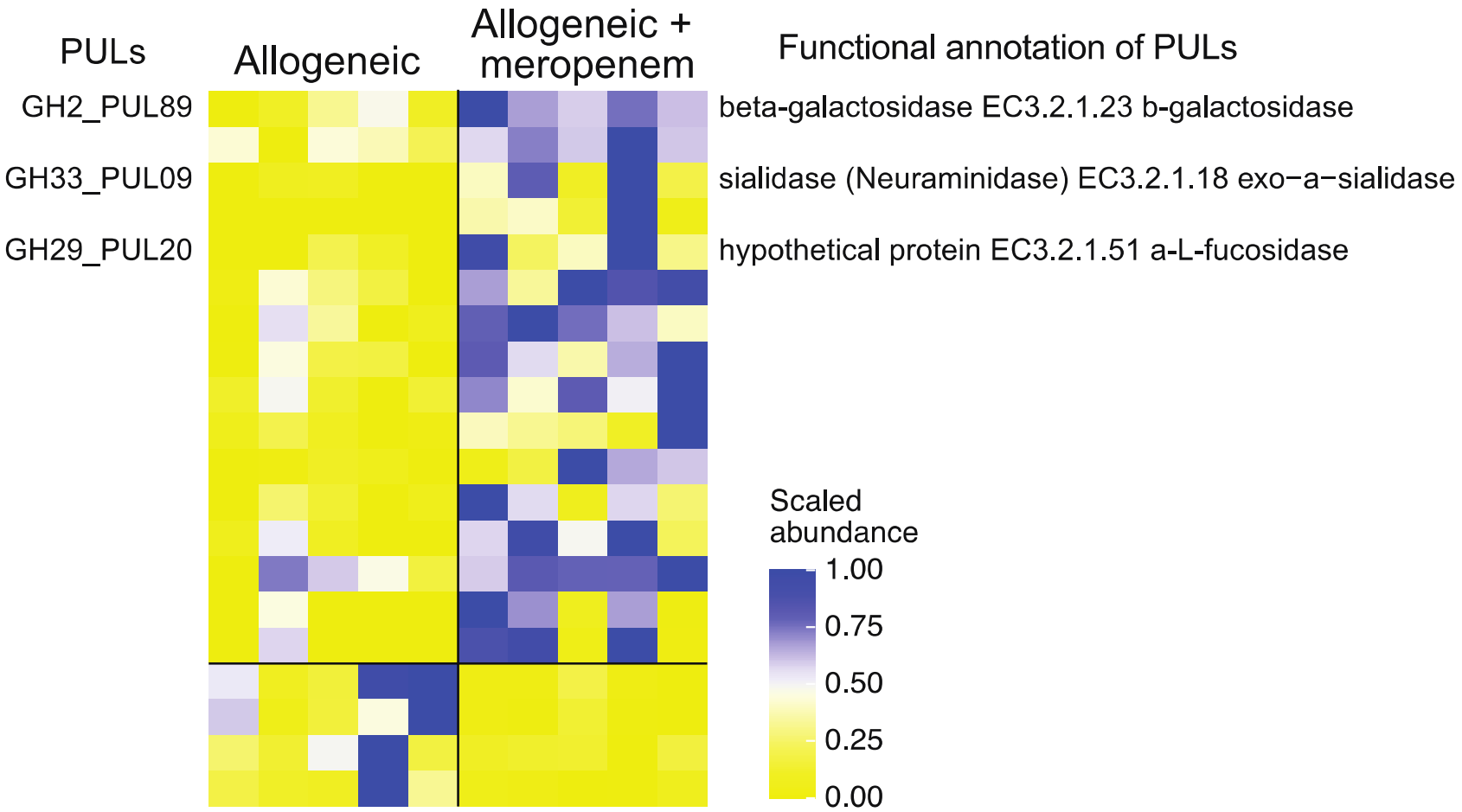


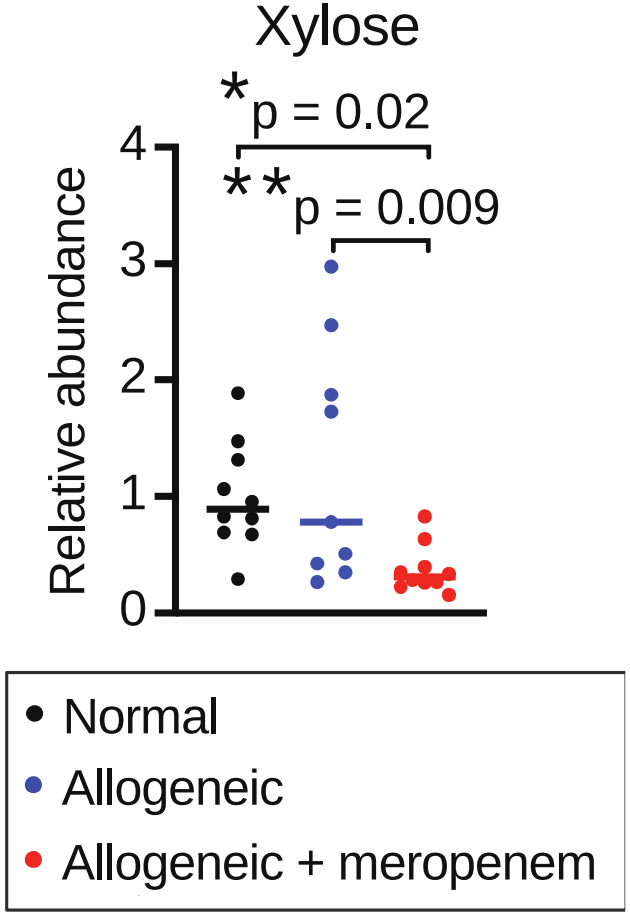
Bars; 100 um

Bacterial translocation is aggravated in mice with GVHD treated with meropenem



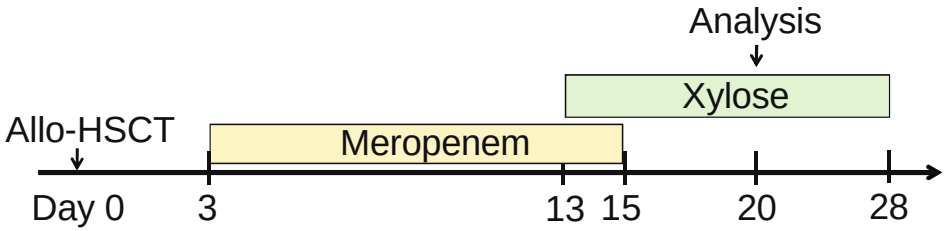
Meropenem changes mucus-degrading enzyme gene expression in *B. thetaiotaomicron*



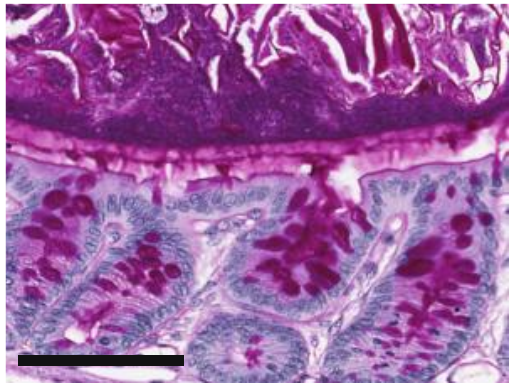


Courtesy of Jamal, Tina, Tomo, Lin, Lucas and Phil

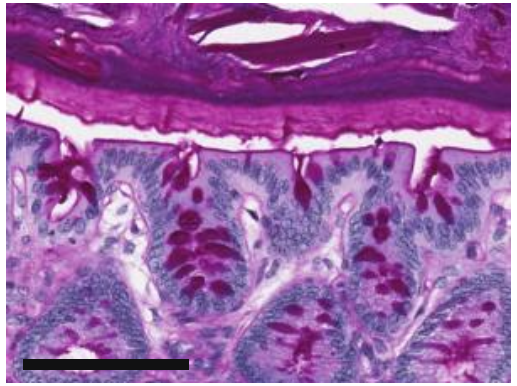
Oral supplementation with xylose benefits mice with GVHD aggravated by meropenem

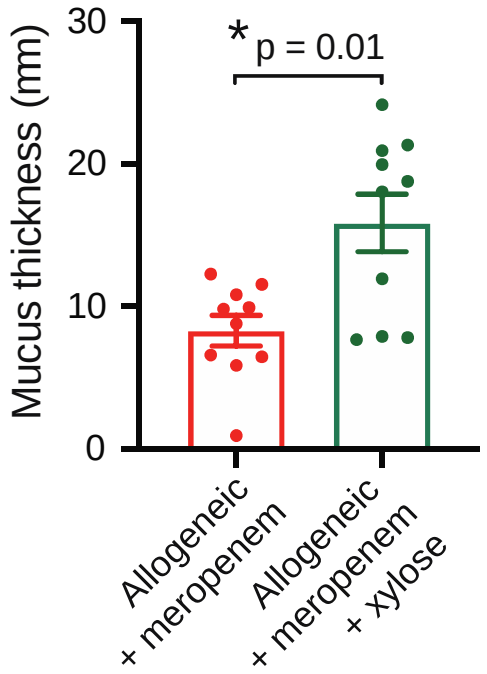


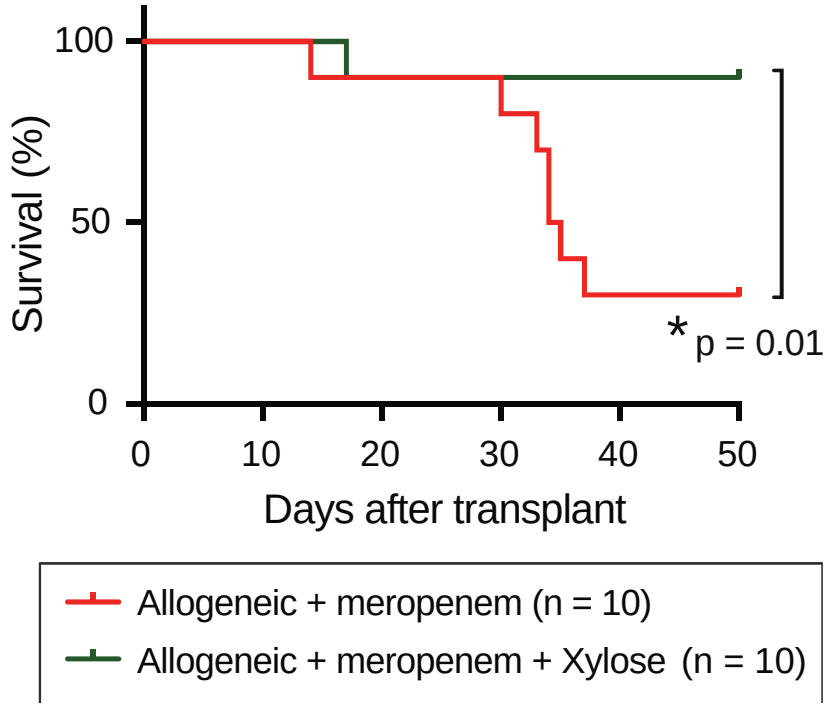
Allogeneic + meropenem



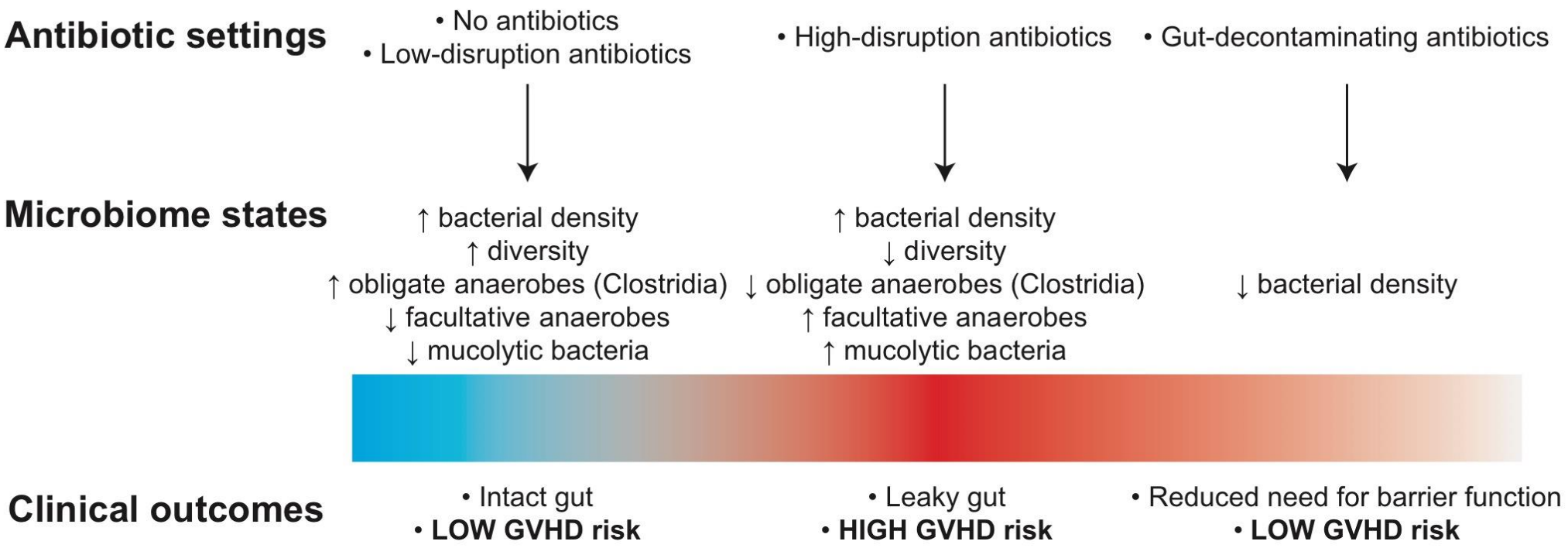
Allogeneic + meropenem + xylose

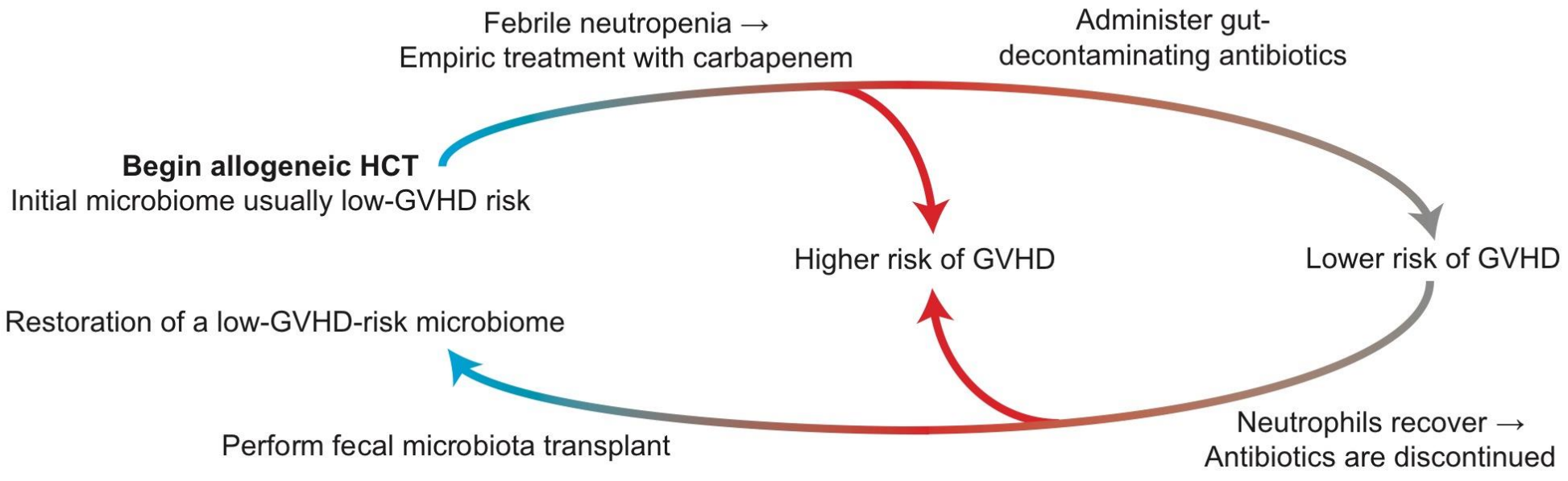






Courtesy of Saira


Antibiotic settings
• No antibiotics
• Low-disruption antibiotics
• High-disruption antibiotics
• Gut-decontaminating antibiotics
Microbiome states
↑ bacterial density
↑ diversity
↑ obligate anaerobes (Clostridia)
↓ facultative anaerobes
↓ mucolytic bacteria
↑ bacterial density
↓ diversity
↓ obligate anaerobes (Clostridia)
↑ facultative anaerobes
↑ mucolytic bacteria
↓ bacterial density
Clinical outcomes
• Intact gut
• LOW GVHD risk
• Leaky gut
• HIGH GVHD risk
• Reduced need for barrier function
• LOW GVHD risk


Febrile neutropenia →
Empiric treatment with carbapenem
Administer gut-decontaminating antibiotics
Begin allogeneic HCT
Initial microbiome usually low-GVHD risk
Higher risk of GVHD
Lower risk of GVHD
Restoration of a low-GVHD-risk microbiome
Perform fecal microbiota transplant
Neutrophils recover →
Antibiotics are discontinued

Acknowledgements

Genomic Medicine
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Christopher Sanchez
Alexandria Brown
Dung (Kate) Pham
Rawan El-Himri
Taylor Halsey
Annette Frenk
Brooke Heckel
Yimei Jin
Wen-bin Tsai
Ivonne Flores
Rishika Prasad
Lauren McDaniel

Genomic Medicine
Andy Futreal
Curtis Gumbs
Latasha Little
Rebecca Thornton
Marcus Coyle
Christigale Mandapat

MDACC
Stem cell transplantation and Cellular Therapy
Amin Alousi
Gabriela Rondon
Jeffrey Molldrem
Richard Champlin
Erizabeth J Shpall

MDACC

Metabolomics Core Facility
Philip L Lorenzi
Lin Tan
Lucas J Veillon

Infectious Diseases
Samuel Shelburne
Roy F. Chemaly
Jessica Galloway-Peña

PRIME-TR
Jennifer A Wargo
Nadim J Ajami

Yale School of Medicine
Chen Liu

University of Michigan
Eric C Martens

CPRIT

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MD Anderson Cancer Center®