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The gut microbiome in pediatric stem cell transplantation: challenges and perspectives

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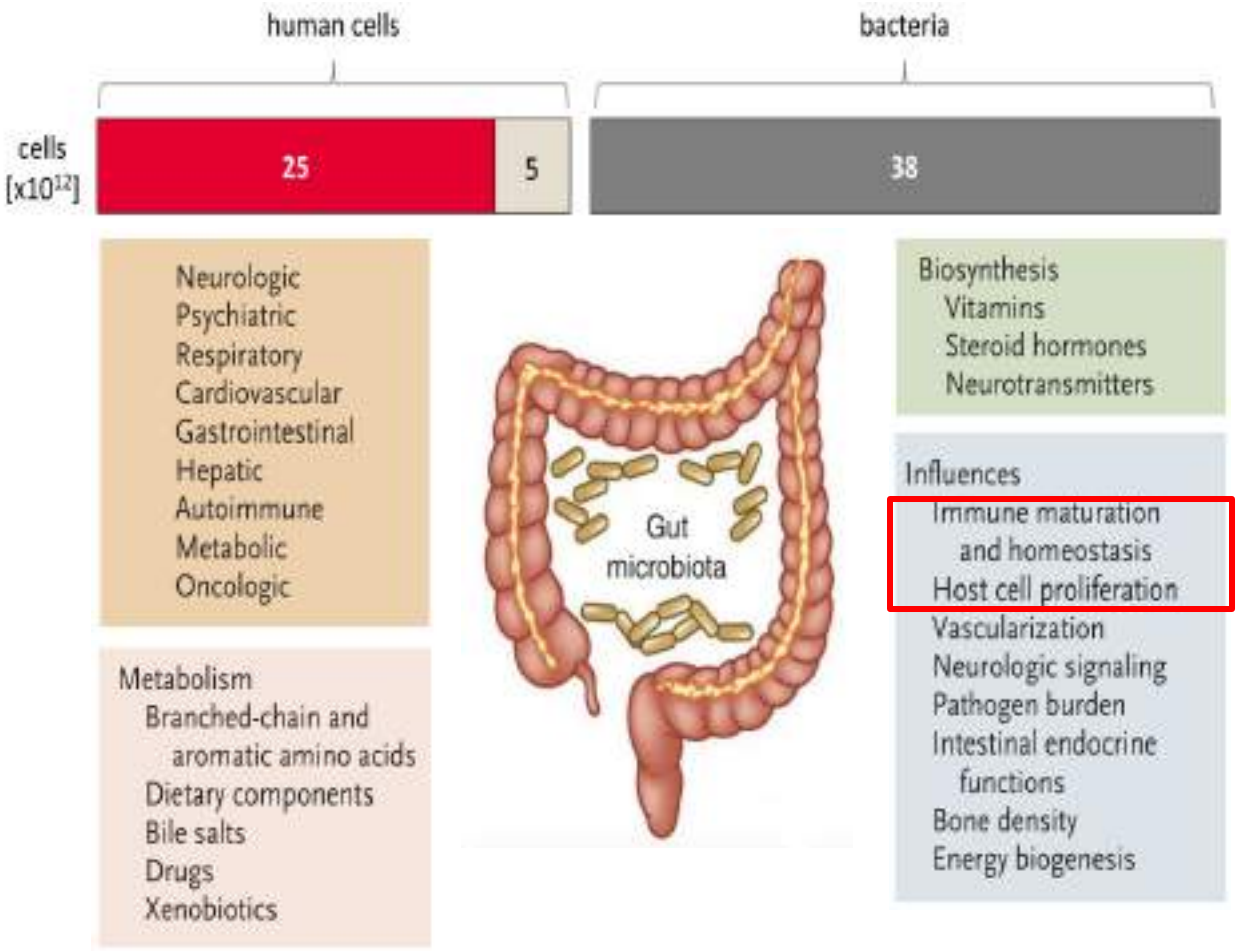
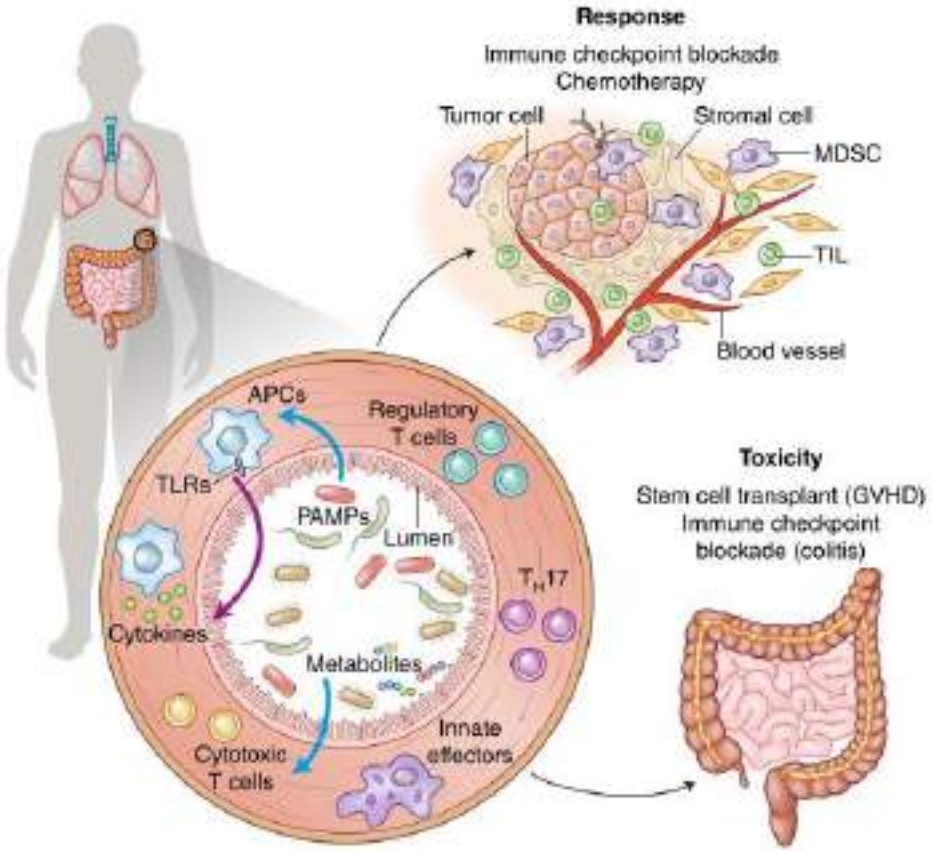
EPTRI General Assembly 13-15 March,2025-Bologna, Italy

The Human Gut Microbiome in Health and Disease





Sender et al., 2016, PLoS Biology

Lynch e Pedersen, 2016, NEJM

Helmink, 2019, Nature Medicine

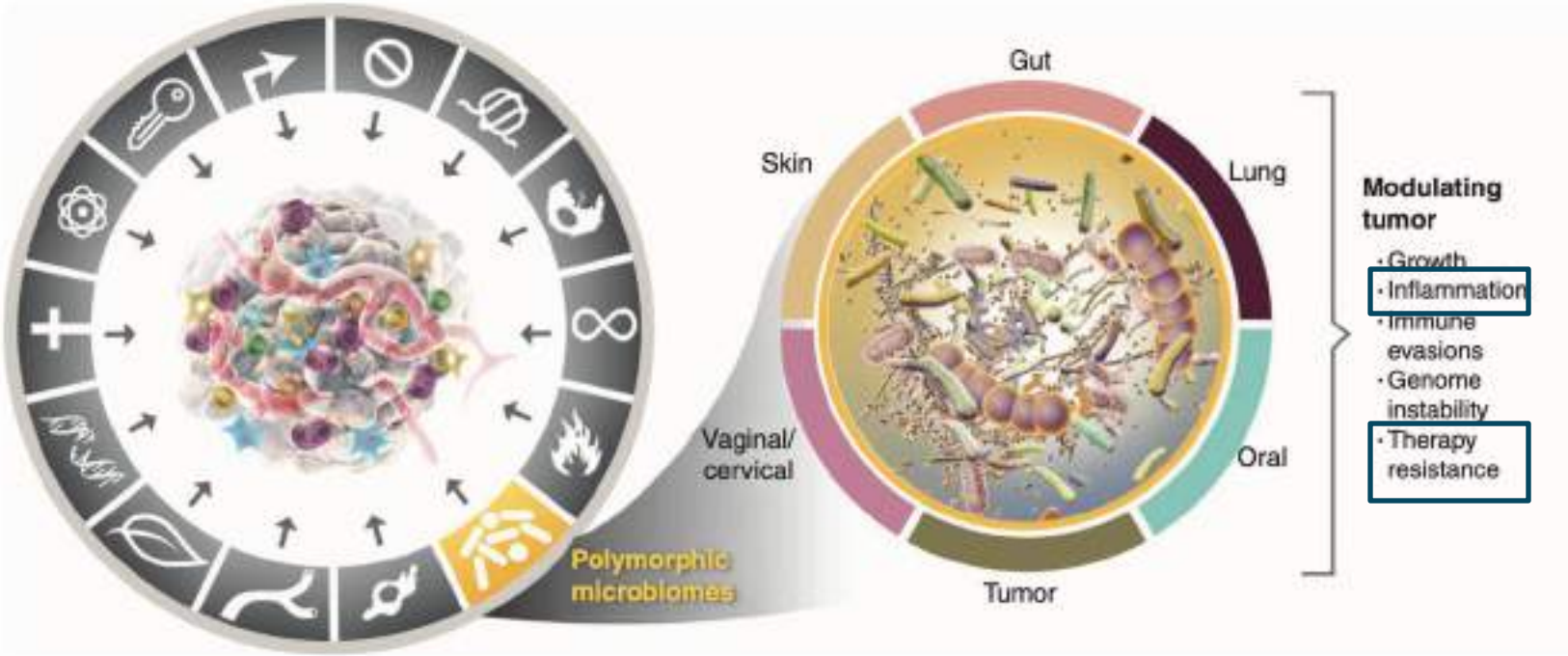
Gut Microbiome and cancers

CANCER DISCOVERY

Hallmarks of Cancer: New Dimensions

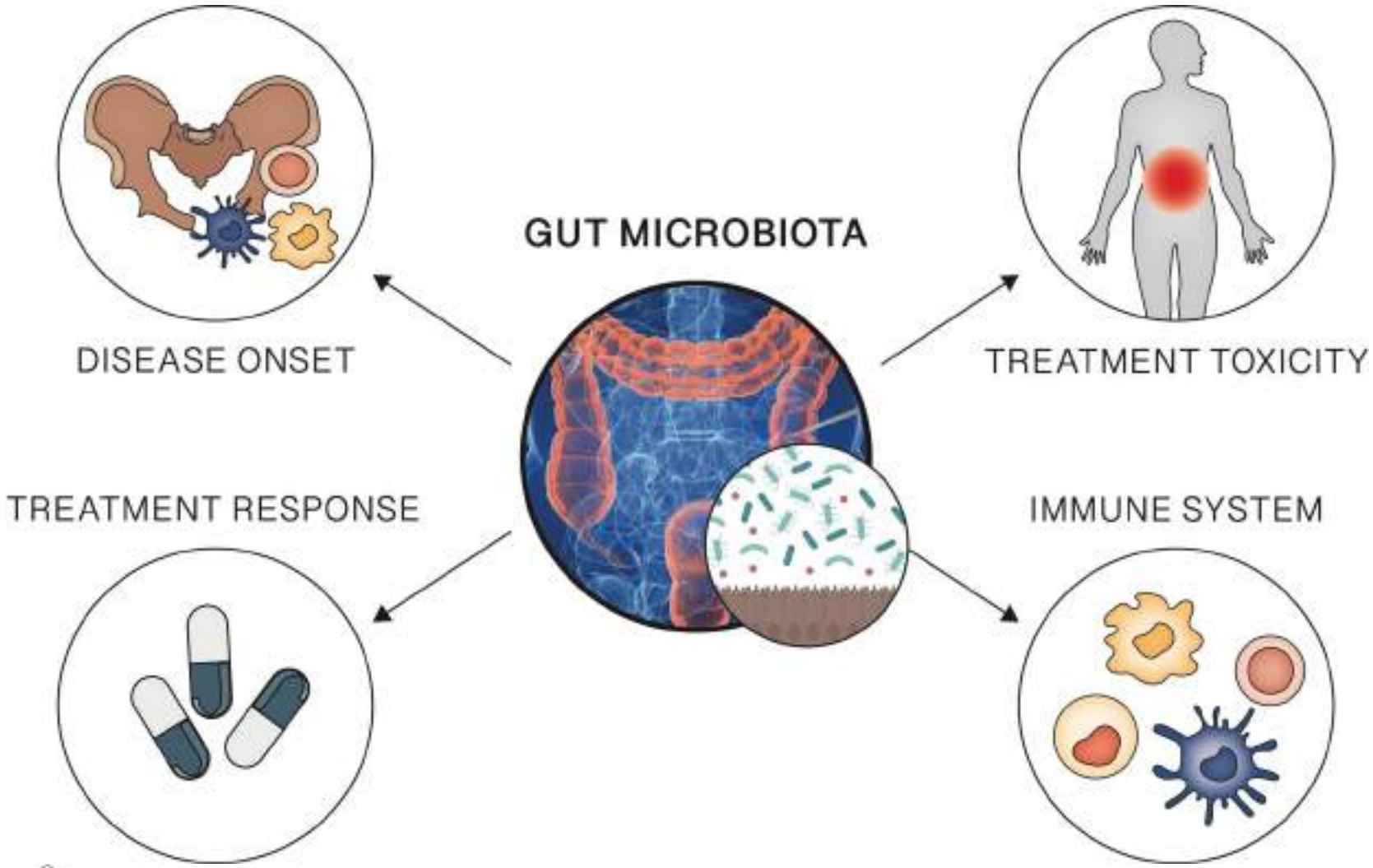
Immunotherapy of Cancer:

- Allogeneic Stem cell Transplantation
- CAR-T Cell Therapy
- Checkpoint Blockade



Hanahan D, 2022 Cancer Discovery

Gut Microbiome and haematological malignancies



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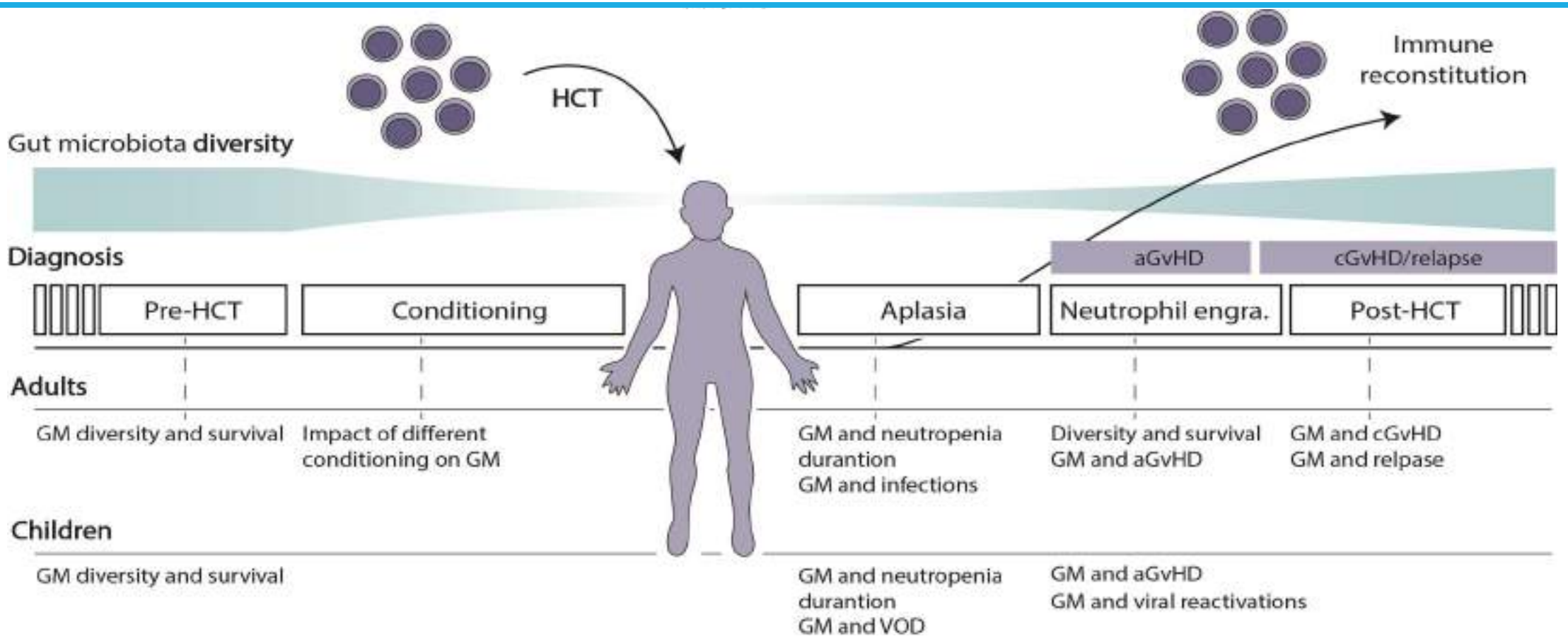
Gut Microbiome and stem cell transplantation



- In adult HSCT recipients, outcomes linked with intestinal bacteria include **overall survival**, **acute and chronic GvHD**, **infections**, **relapse**, and **immune reconstitution.**
- Data regarding children undergoing allo-HSCT have thus far been lacking, but some of these associations have been described in smaller cohorts.

Shono et al., 2018, Nature Reviews

Gut Microbiome and stem cell transplantation

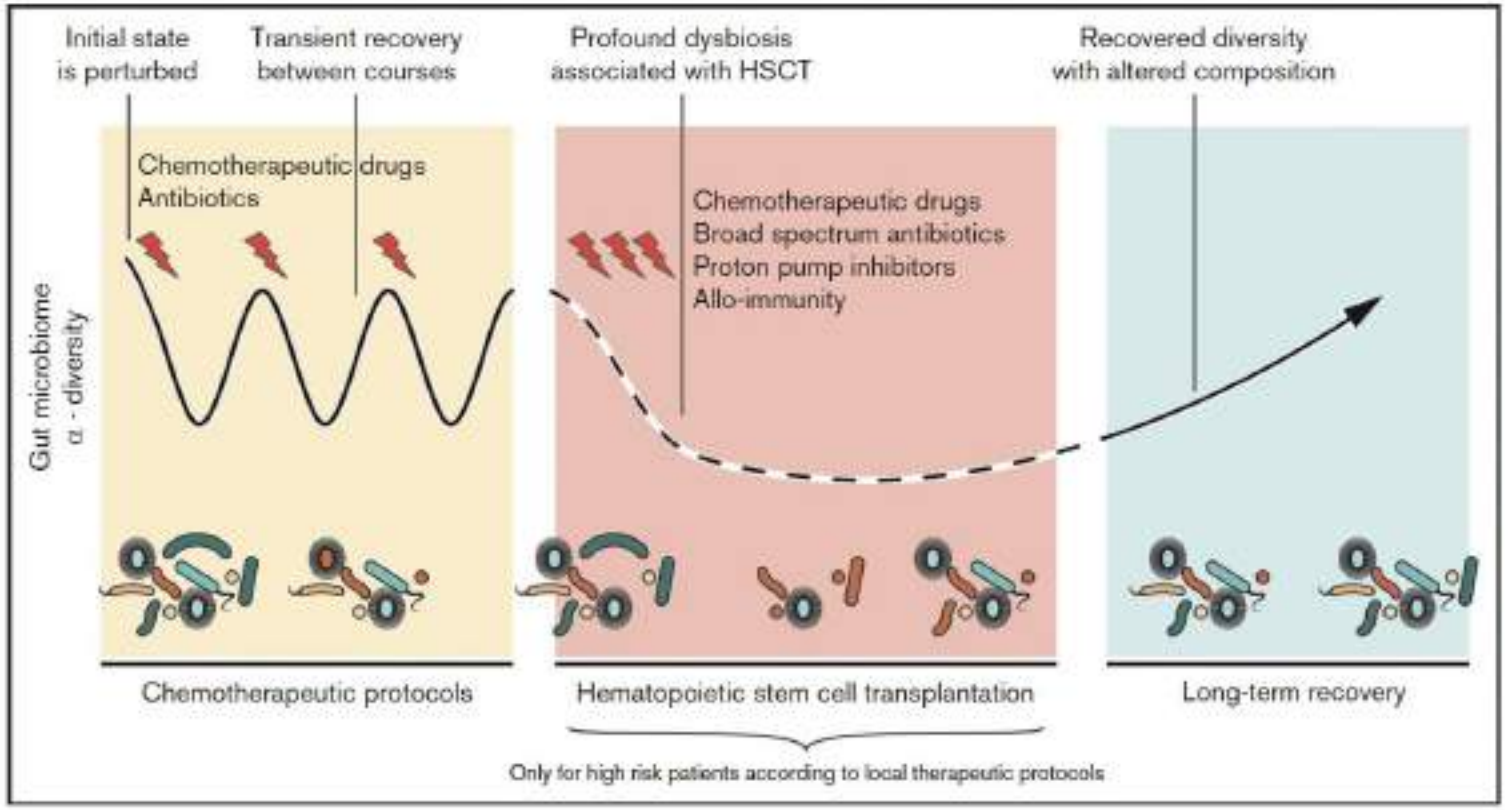


Figure 2. The trajectory of the GM during the therapeutic course of AL. The initial microbial state is perturbed by chemotherapeutic cycles with partial recovery between them. HSCT exerts a strong dysbiotic effect on the GM. Reconstitution after HSCT resembles the pre-HSCT state, but dysbiotic features often persist.

HSCT produces a **loss of diversity** of the intestinal microbiome in particular **commensal anaerobes** (taxa affiliated with the order Clostridiales, eg. Lachnospiraceae and Ruminococcaceae) and a **shift towards an enteropathogenic flora with a predominance** of Gram-negative Enterobacteria (E. coli, Klebsiella, Enterobacter spec.) and Gram-positive Lactobacillales (Lactobacillus, Enterococcus and Streptococcus spec.) during the course of the transplant.

After about **2-3 months the ecosystem recovers** its initial richness and metabolic capacity albeit with persistent dysbiotic features.

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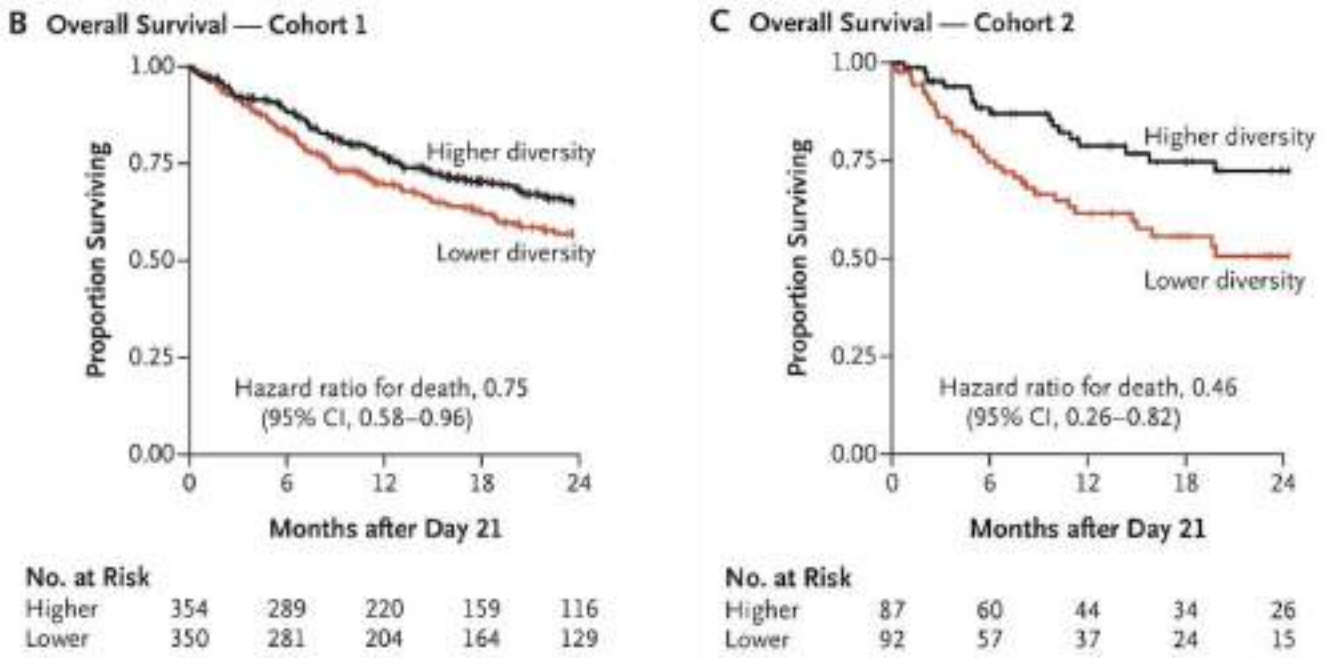
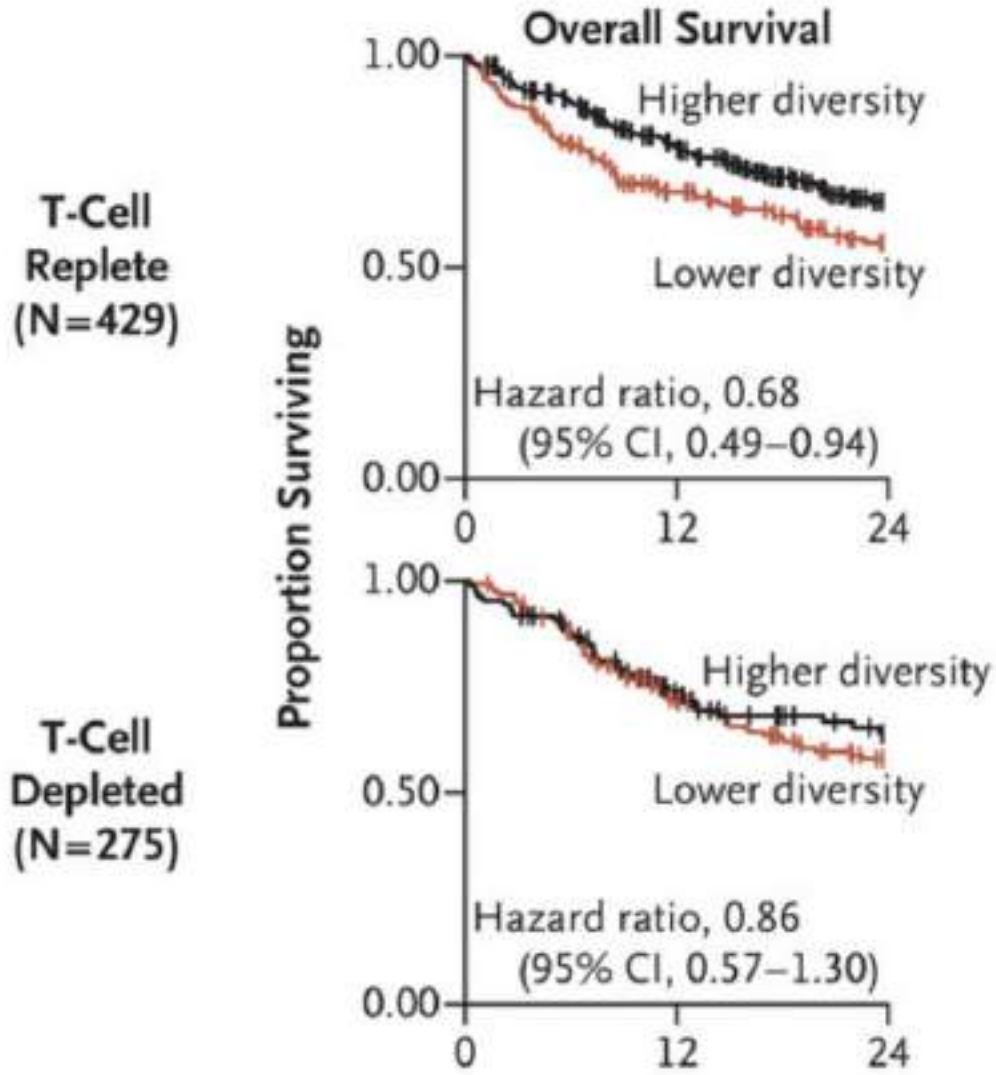
Masetti et al., 2021, *Blood Adv*

Gut Microbiome diversity predicts survival in stem cell transplantation



- Overall survival was longer among patients with higher **intestinal diversity in periengraftment** samples.
- This association was observed among recipients of unmodified grafts and not among recipients of **T-cell–depleted grafts**.



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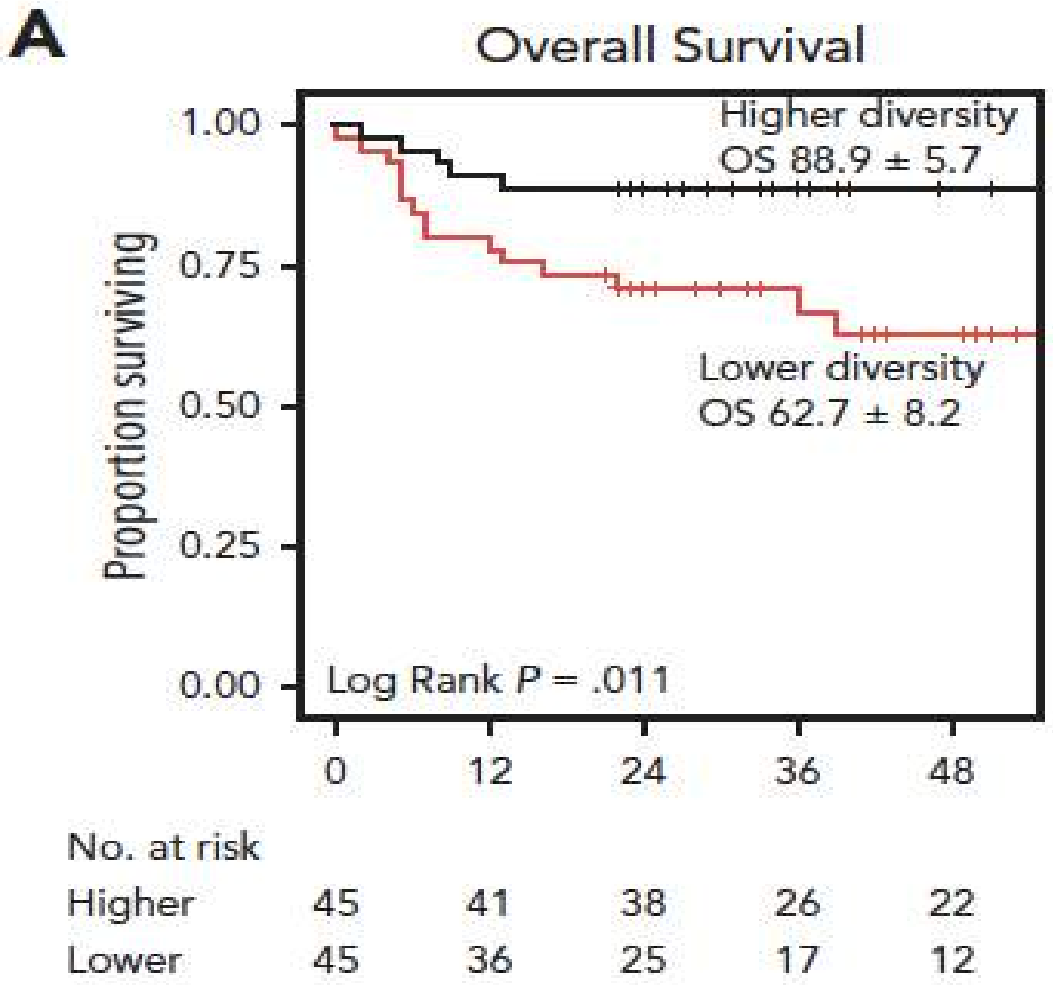
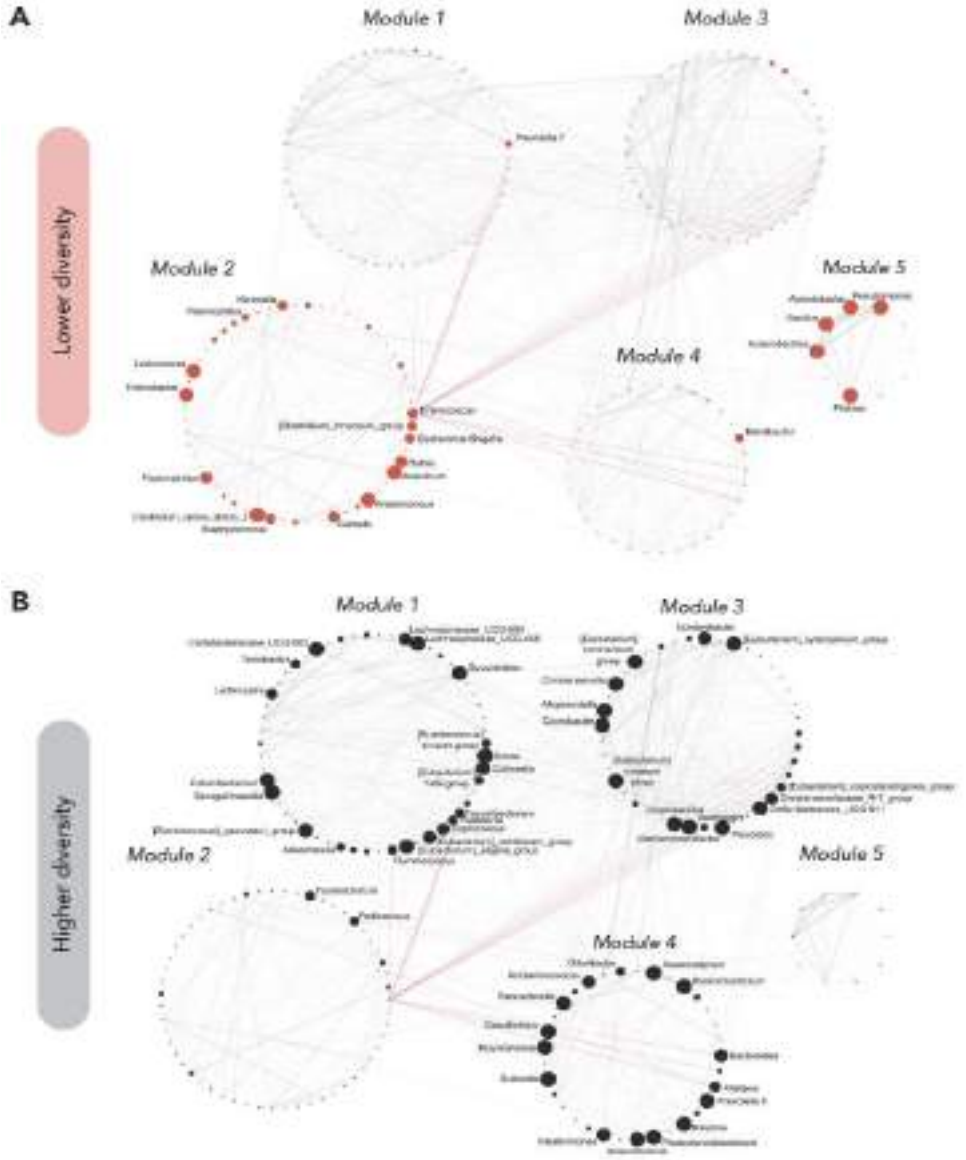
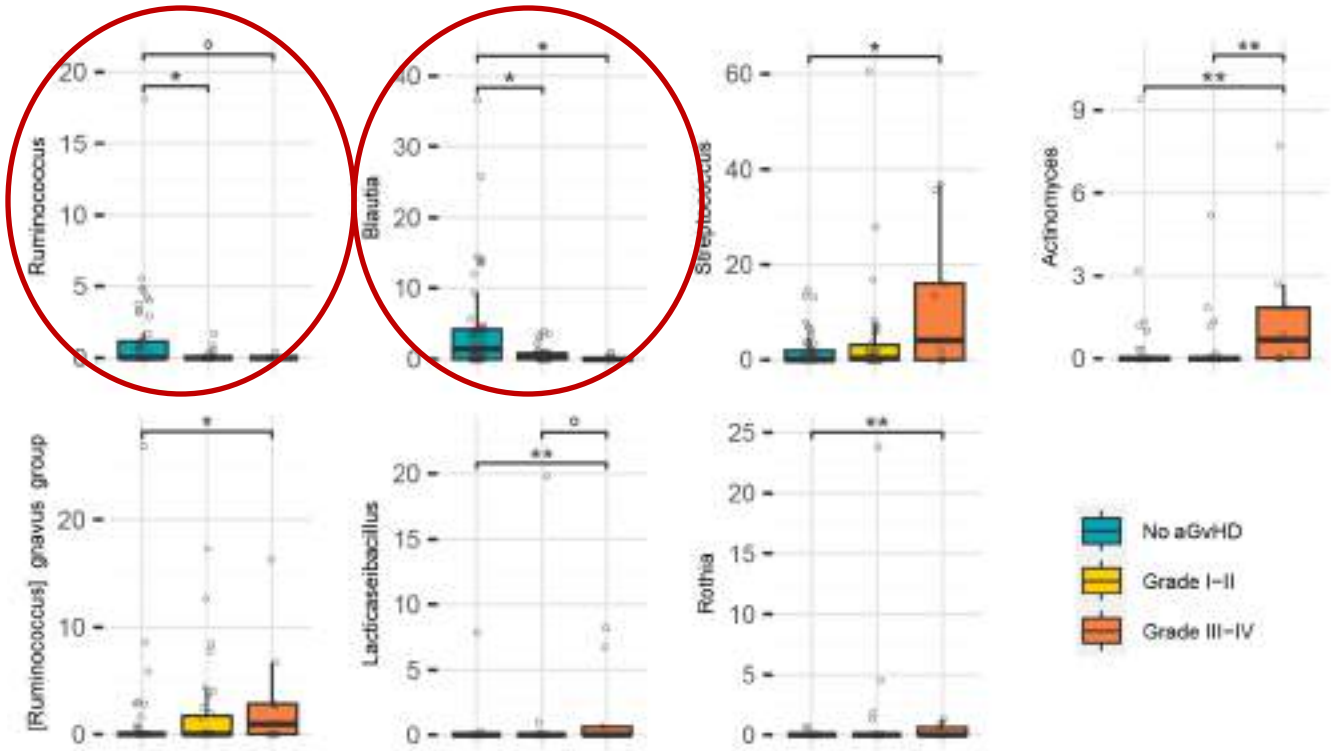
Gut Microbiome diversity predicts survival in stem cell transplantation

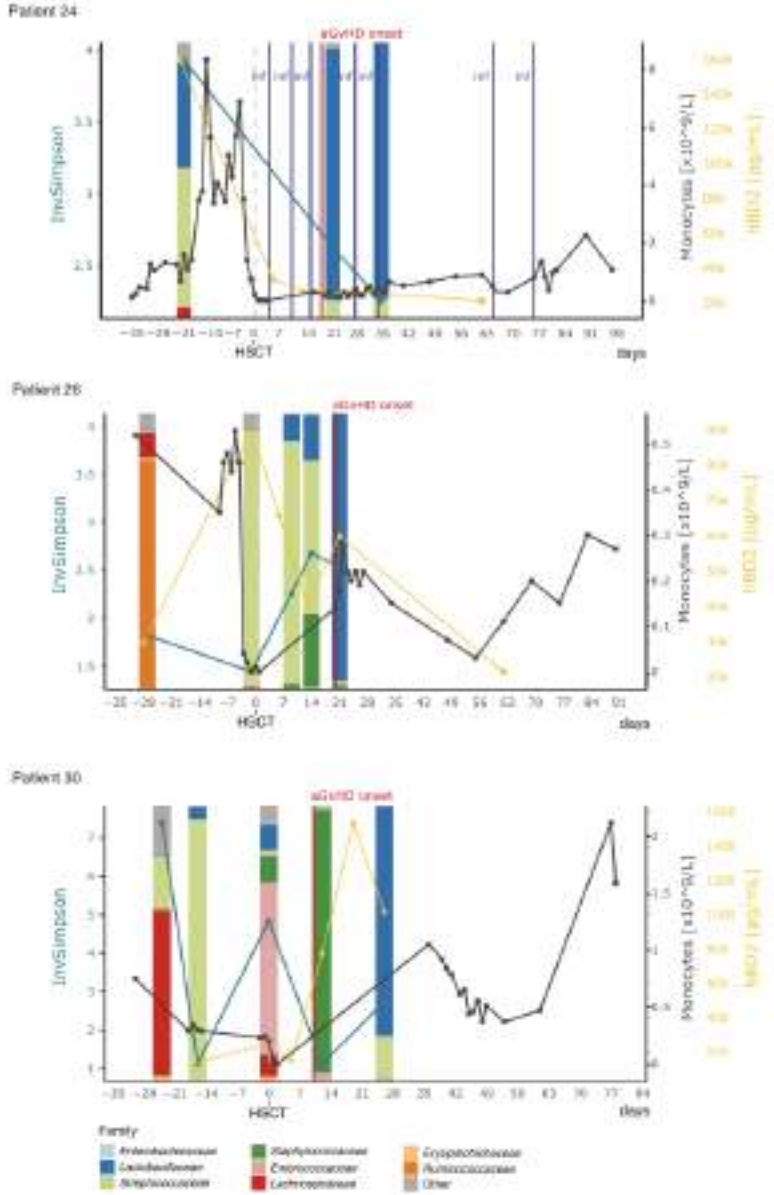




Masetti et al., 2023, Blood

GM diversity pre-SCT protects from GvHD development and mortality





Higher pre–allo-HSCT relative abundances of Blautia and Ruminococcus appeared to be protective against the subsequent aGVHD development. Abundances of Lactobacillaceae increased predominantly at the aGvHD and are predictive of GvHD mortality.

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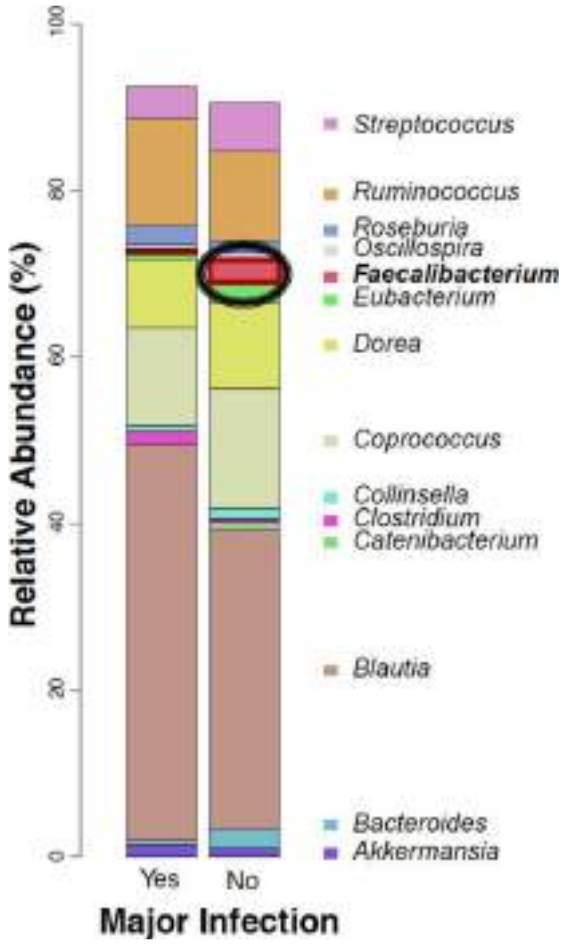
Masetti et al., 2023, Blood; Ingham et al, 2021, Microbome

GM domination and specific GM signatures are associated with BSI



Expansion of *Faealibacterium* in the baseline GM was predictive of protection from major bacterial infection.

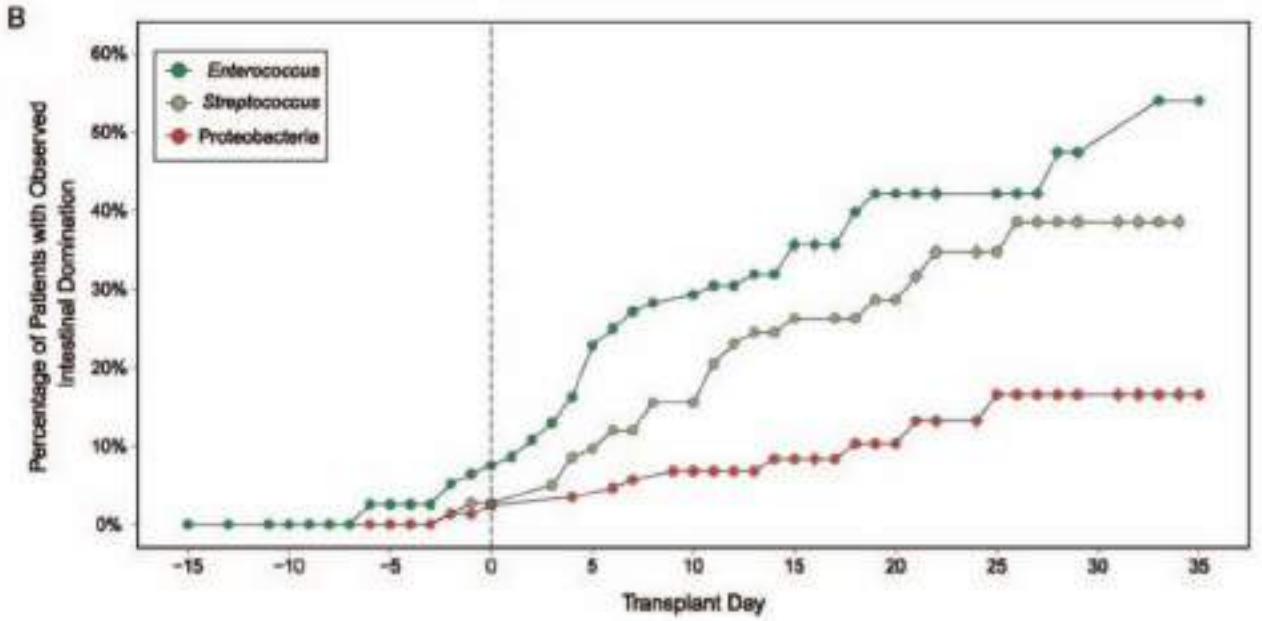


Table 3. Association of Intestinal Domination With Bacteremia[a]

	VRE Bacteremia		Gram-negative Bacteremia	
Dominating Taxon[b]	HR (95% CI)	*P*	HR (95% CI)	*P*
Enterococcus	9.35 (2.43–45.44)	.001	1.35 (.25–5.08)	.690
Streptococcus	0.21 (.00–1.75)	.184	0.82 (.09–3.65)	.823
Proteobacteria	0.75 (.01–6.14)	.837	5.46 (1.03–19.91)	.047

Intestinal domination (occupation of at least 30%) correlated with subsequent development of a correspondent BSI with VRE or Gram- bacteria

Murthy et al., 2020, *BBMT*

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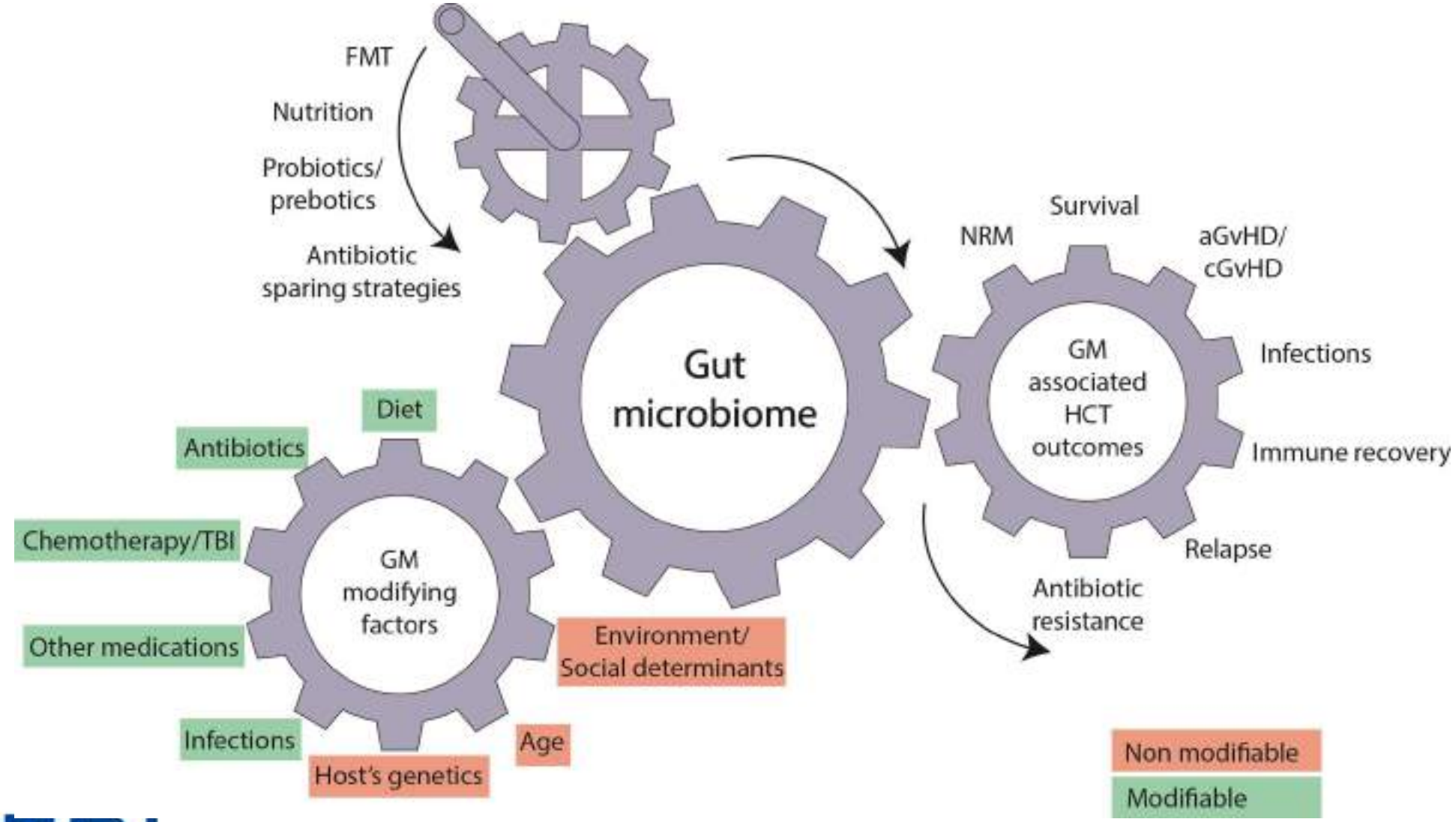
How can we turn the tide?



Trying not to lose homeostasis

«Homeostasis holds complex systems together invisibly; we notice only its failures».

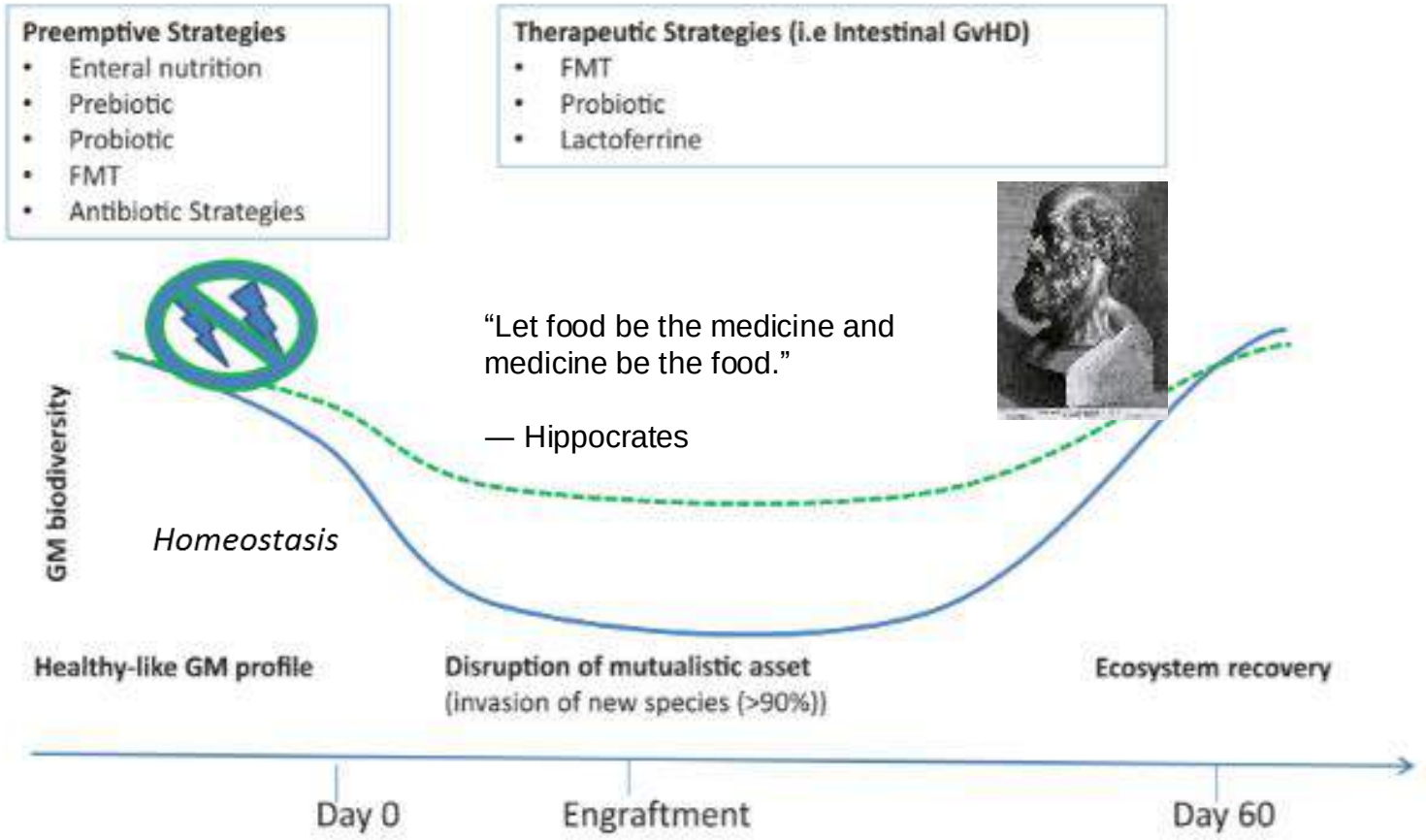


Figure 2. Potential strategies, preemptive and therapeutic, used for preventing or treating GM dysbiosis during HSCT are summarized.
GM, gut microbiota; HSCT, hematopoietic stem-cell transplantation.

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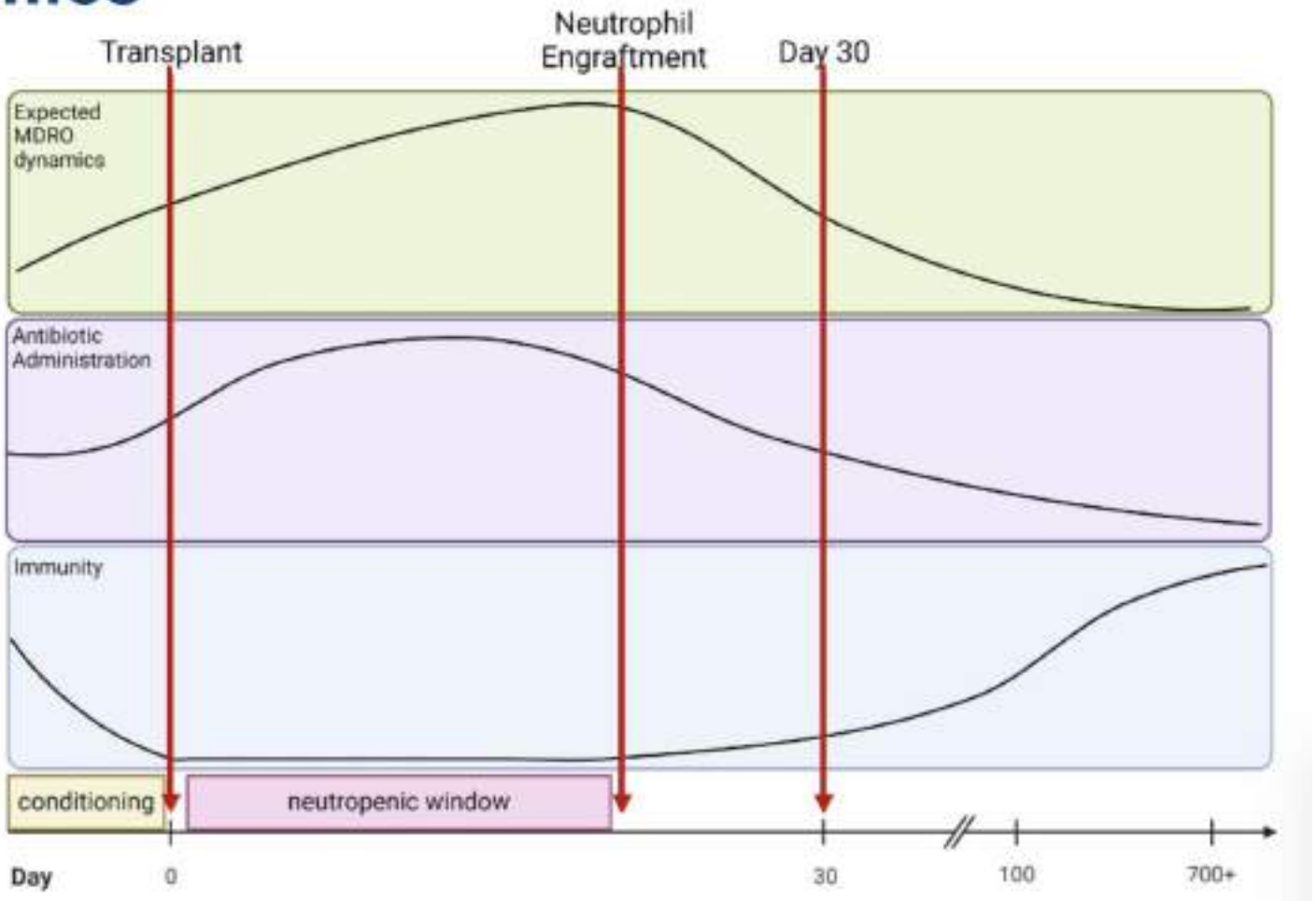
Zama D and Masetti R. Bone Marrow Transplant. 2017

MDRO dynamics determined by metagenomic approach

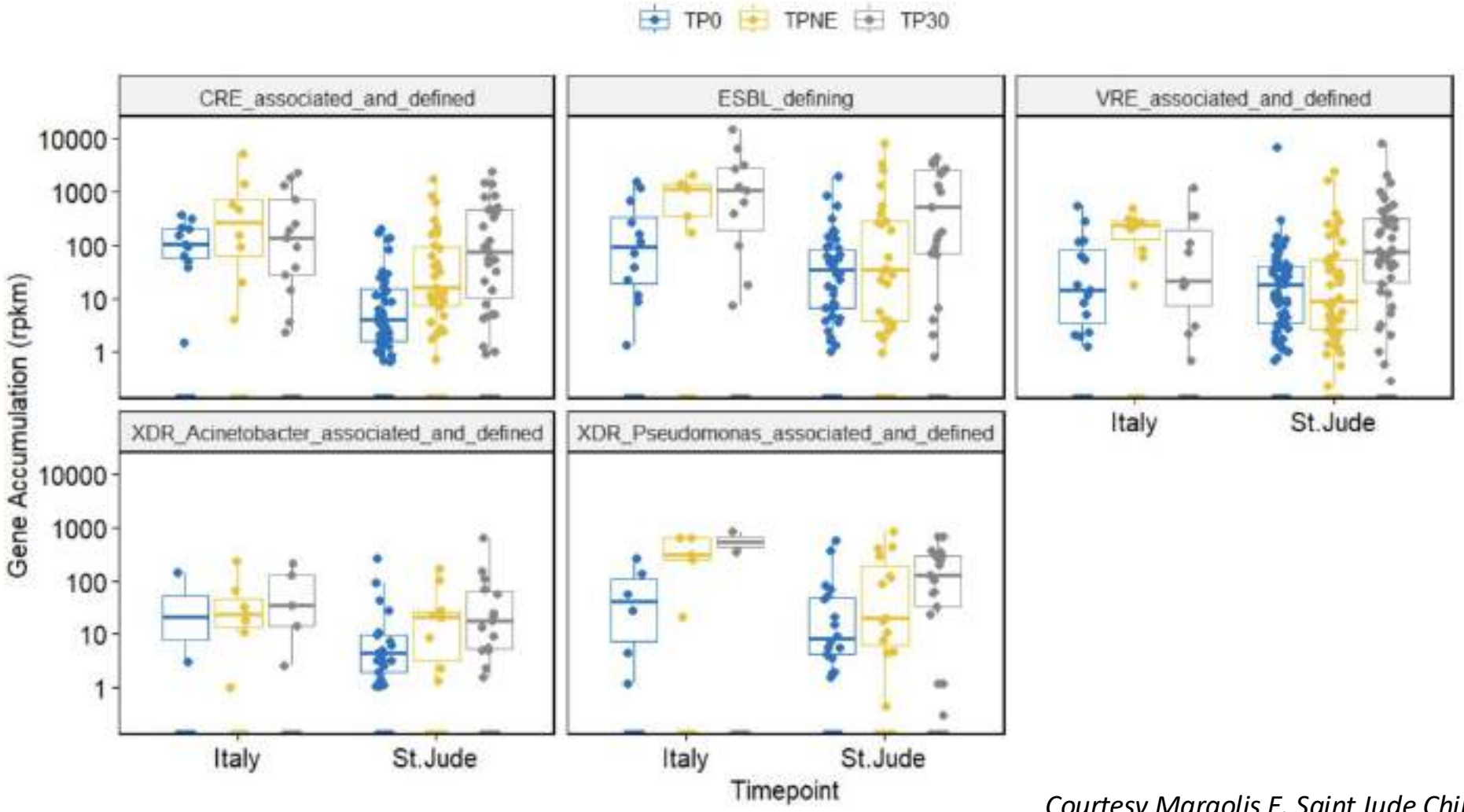
Pediatric HCT patients present a unique opportunity to study a discrete period of heavy antibiotic use and resulting MDRO dynamics

- Combining the low immunity and high rates of antibiotics means the opportunity for MDROs to bloom within the patient microbiomes and potentially lead to infections
- Based on the accepted tenets of antibiotic resistance we could expect MDRO-associated genes to peak in the neutropenic window and then decrease.



MDRO dynamics determined by metagenomic approach

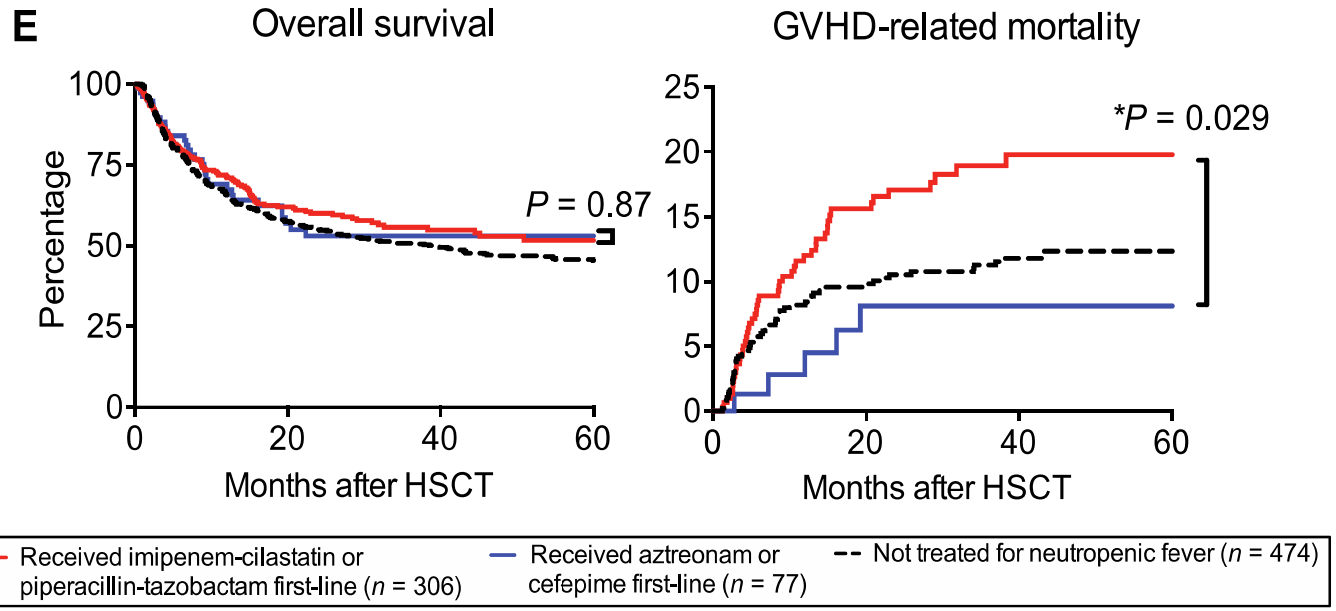
MDRO-associated Genes Continue to Increase to Day 30



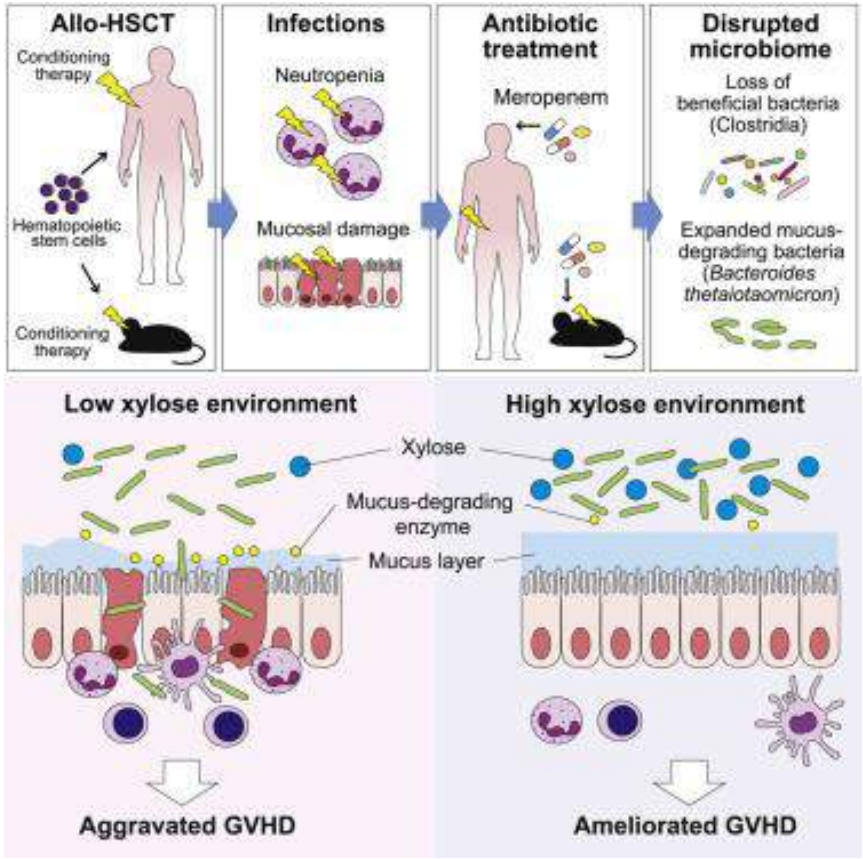
Courtesy Margolis E. Saint Jude Children Hospital, Memphis

Anaerobe-targeting antibiotics increase GvHD mortality by loss of the protective mucus

Treatment of neutropenic fever with **piperacillin- tazobactam and imipenem-cilastatin** was associated with increased GVHD-related mortality, while treatment with aztreonam and cefepime was not.



In mouse models, treatment with **anaerobe targeting antibiotics** was associated with *Akkermansia Muciniphilia* and *Bacteroides Thetaiotomicron* **expansion**, two well known mucin degrader species, loss of mucus layer and worse GvHD severity



Shono et al., 2016, Sci Transl Med ; Hayase et al. 2022, Cell

EN promotes GM homeostasis and reduces transplant complications

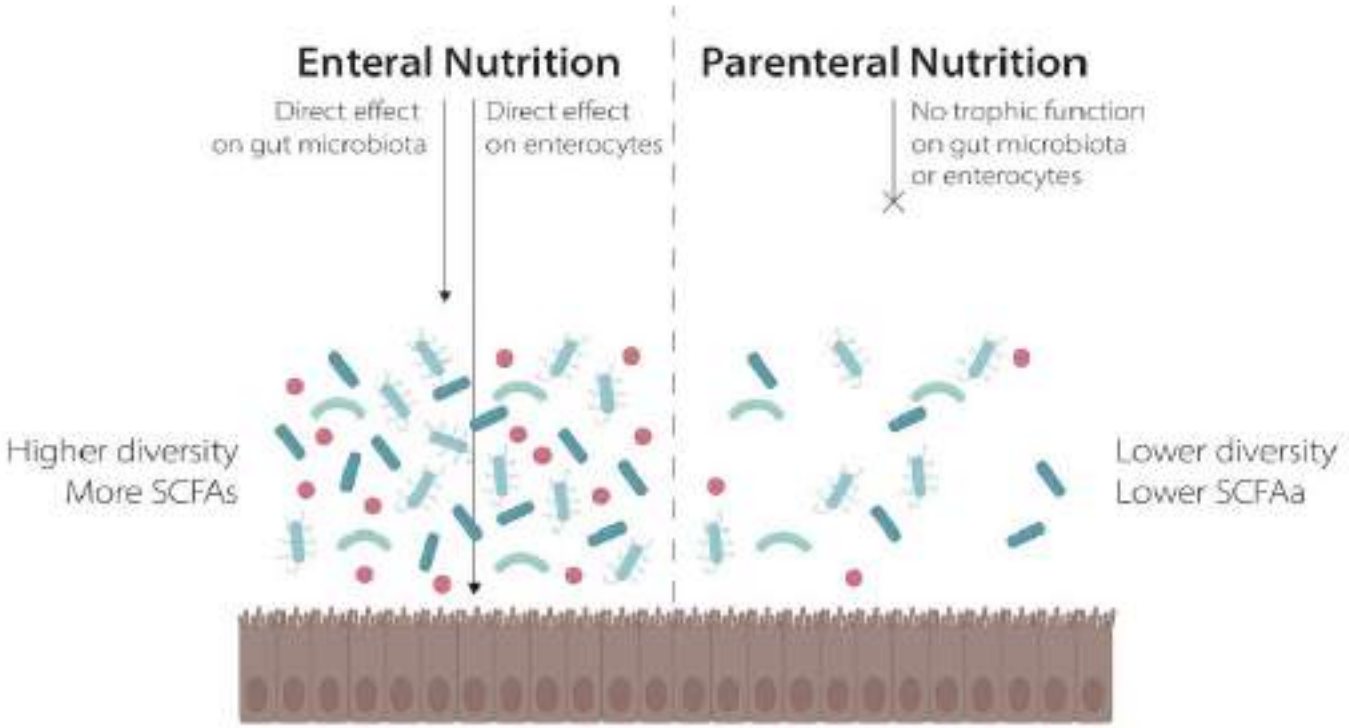
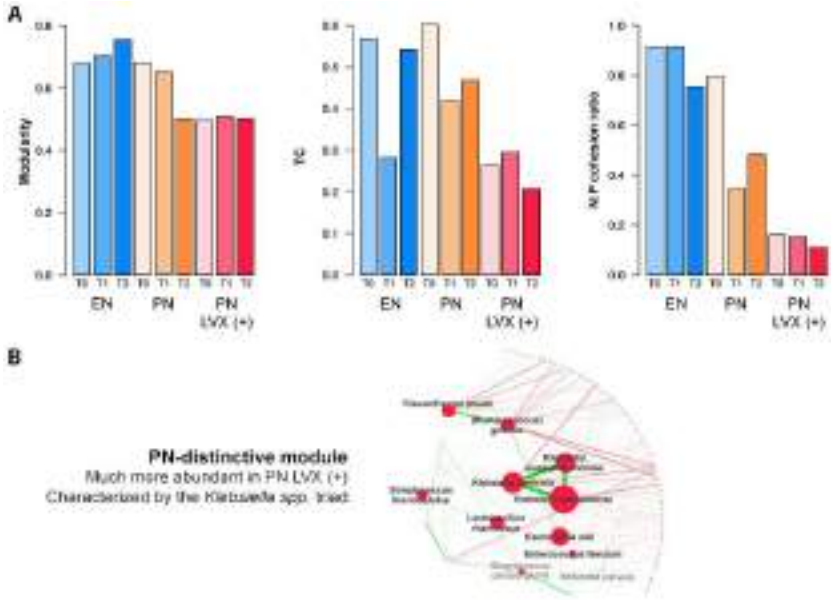


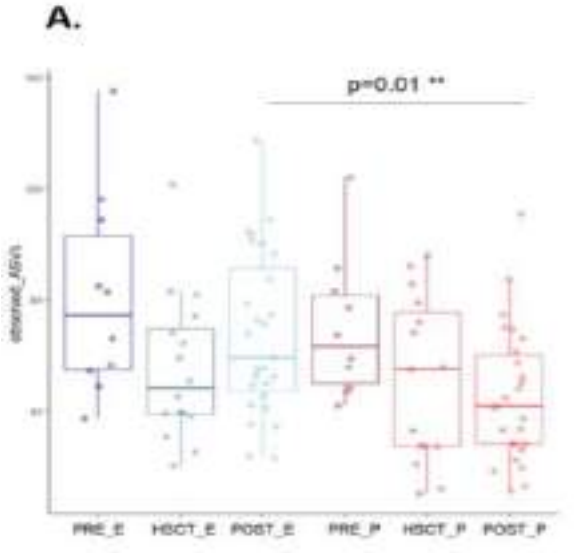
FIGURE 1
The effect of Enteral and Parenteral Nutrition on the gut ecosystem.

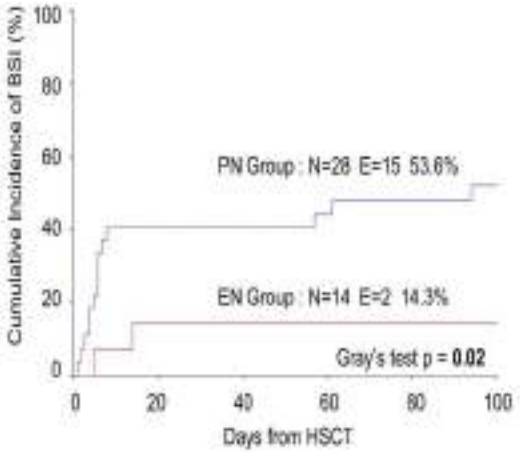
Meratore E, Masetti R et al. Front. In Nutrition 2022

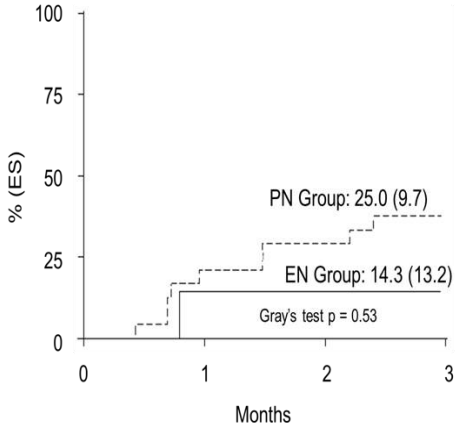
EN promotes GM homeostasis and reduces transplant complications

By evaluating the network topology, we found that **PN, especially preceded by LVX prophylaxis**, resulted in a detrimental effect over the GM, with low modularity, poor cohesion, a **shift in keystone species and the emergence of modules comprising several pathobionts**, such as Klebsiella spp., [Ruminococcus] gnavus, Flavonifractor plautii and Enterococcus faecium.







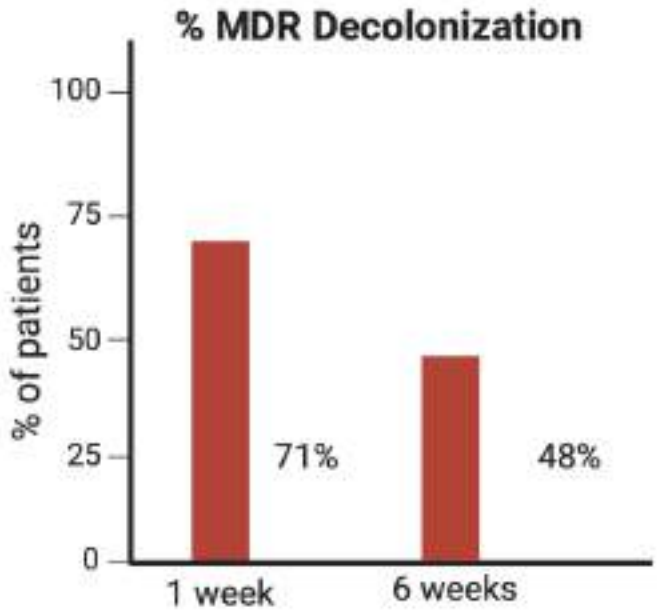


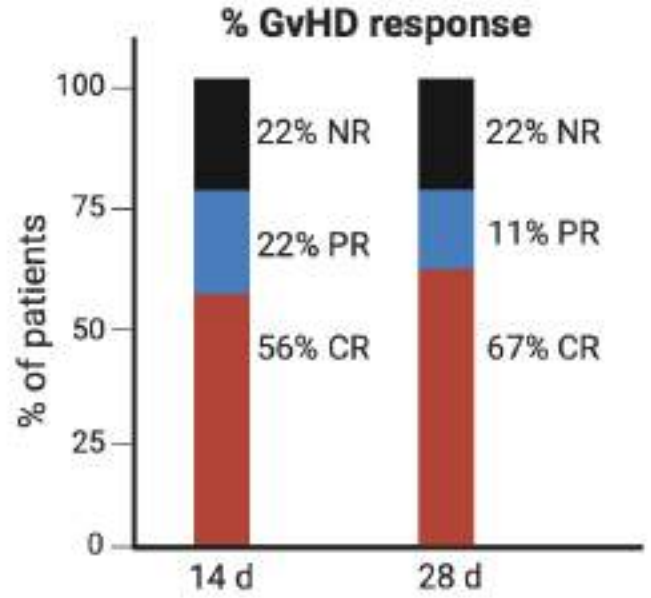
Masetti et al., 2022, Front Nutr; Fabbrini et a., 2023 Comm Biology; Zama et al, 2019 Nutr Journ

Fecal microbiota Transplantation: the Bologna-Roma-Padova experience

FMT infused via **Upper GI**, a median of **150 ml** of fecal material from unrelated donors

	Patients n=28/ infusions n=45
Center Rome/Bologna/Padova	16/9/3
N°. of infusions per patient, median (range)	1 (1-4)
Age, median (range)	4,0 (0,8-18,6)
Type of HCT Haplo/MUD/Sibling/CB, n	11/9/6/2
Indication SR-GvHD/MDR decolonization/both, n	5/18/5
N.° previous lines GvHD therapy, median (range)	4,5 (3-7)
SAE related to FMT, n	0
AE related to FMT n grade	5 I,I,I,II,II





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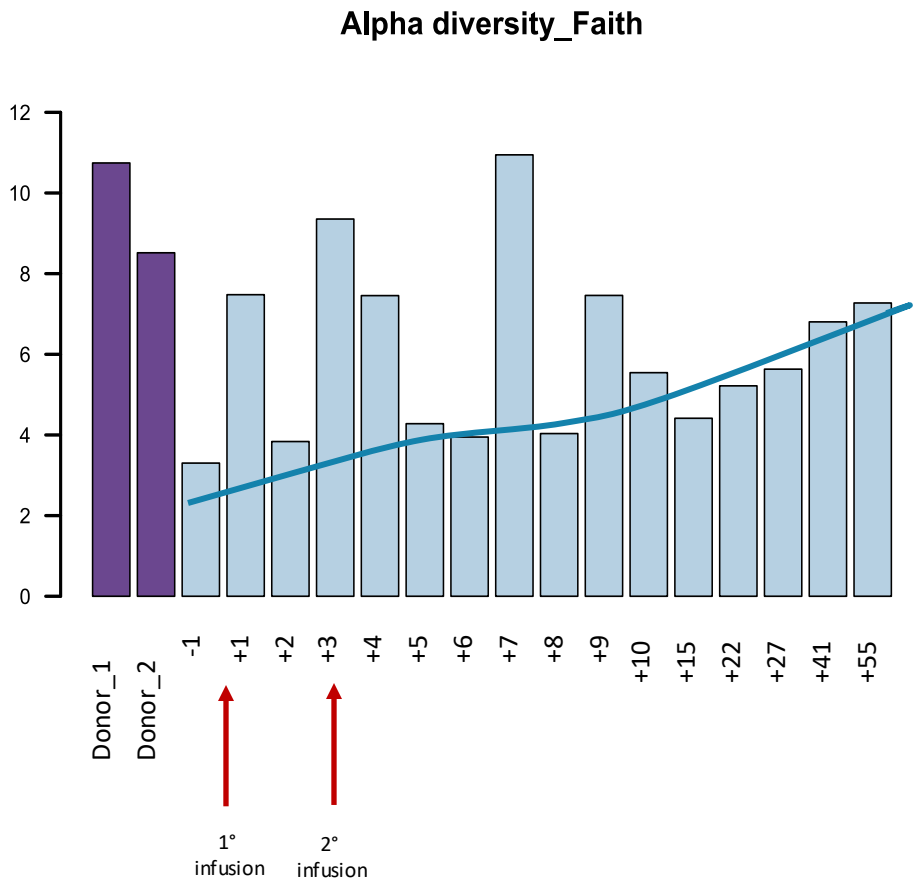
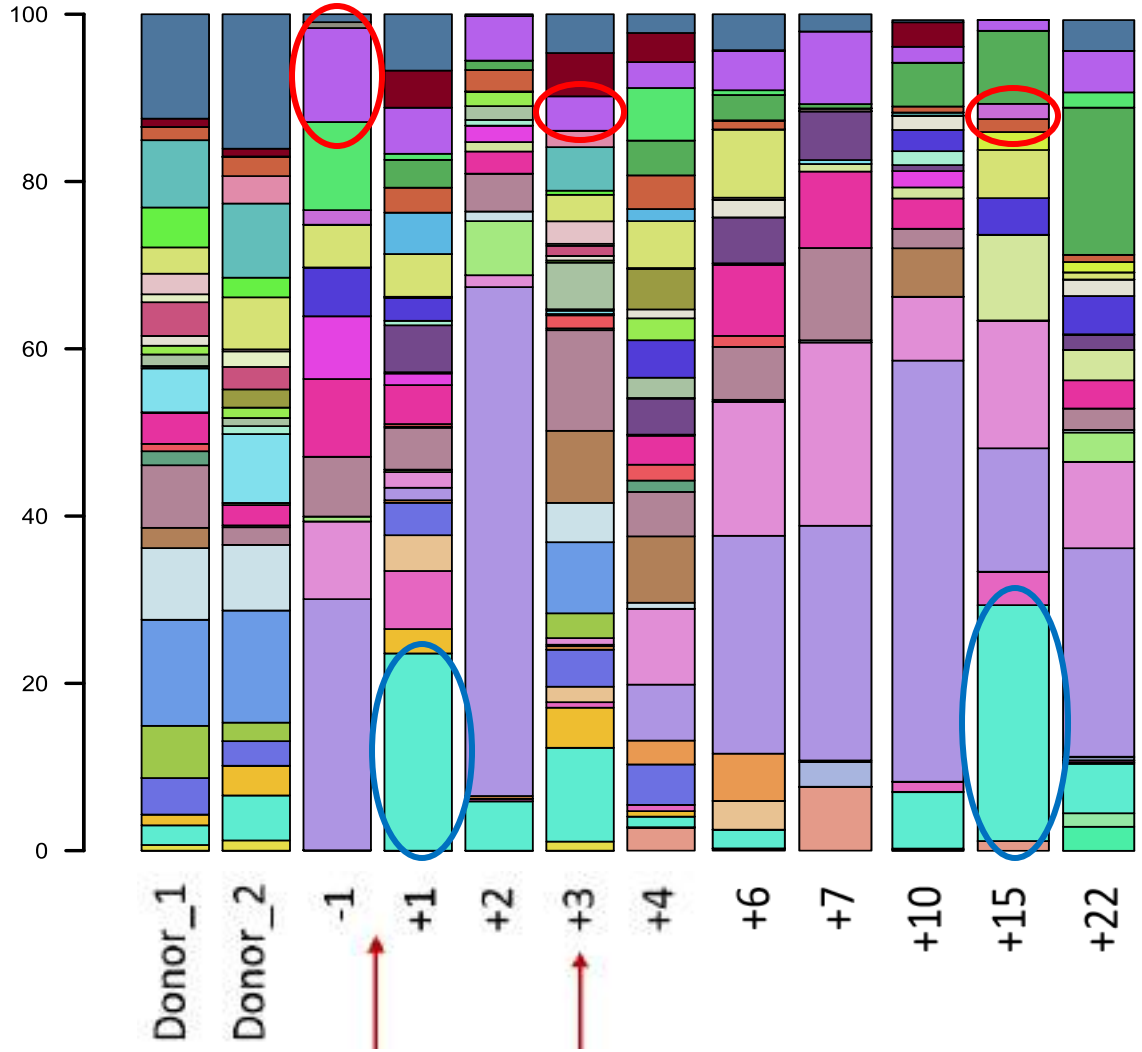
Fecal microbiota Transplantation: the Bologna experience

Alpha diversity slowly and progressively increased after the two infusion. At the same time, enrichment of commensals and loss of pathobionts was observed





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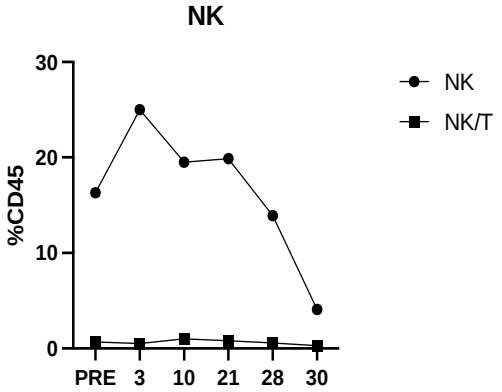
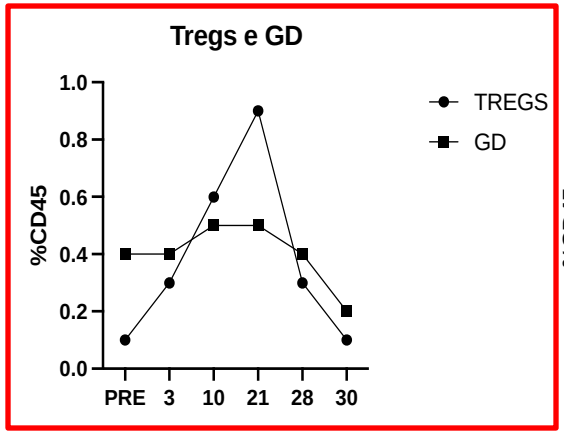
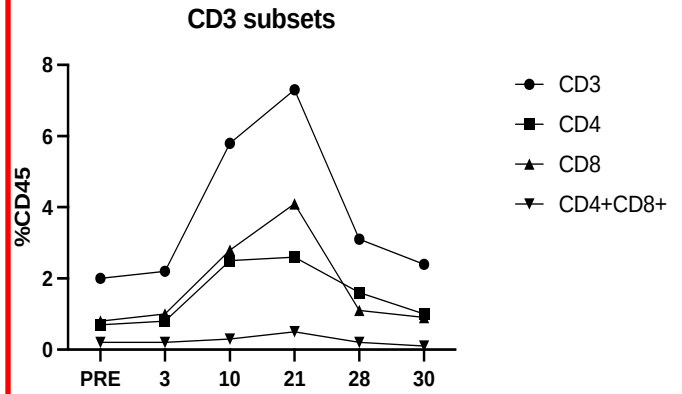
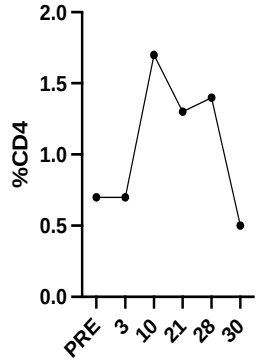
Fecal microbiota Transplantation: the Bologna experience

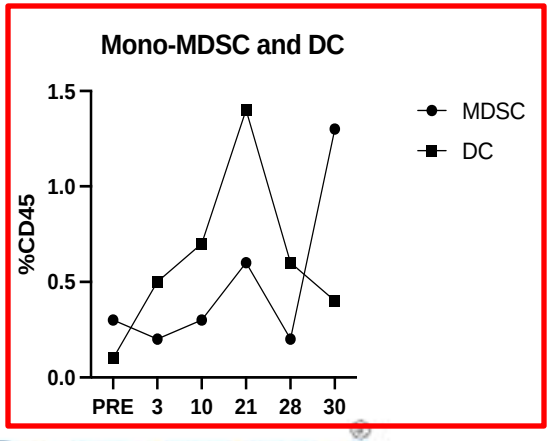
T-regolatory, Monocytic Myeloid-derived suppressor and dendritic cells expansion up to day +21 from FMT, markers of increased immuno-tolerance

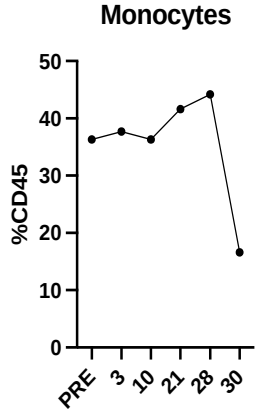


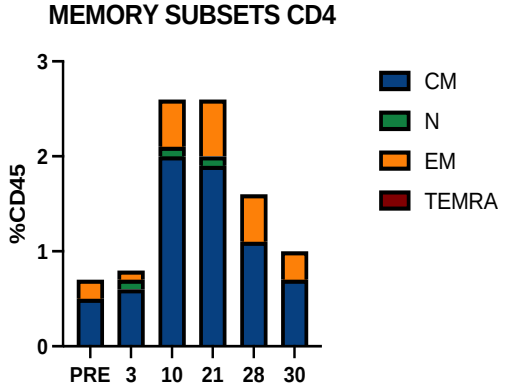


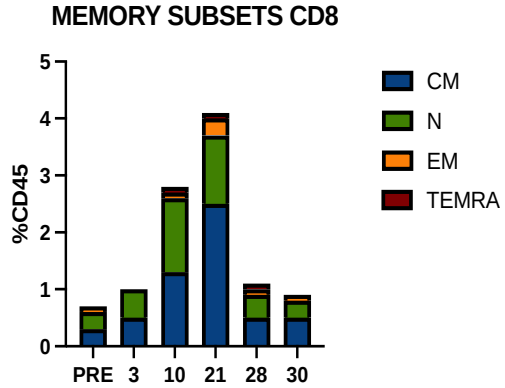












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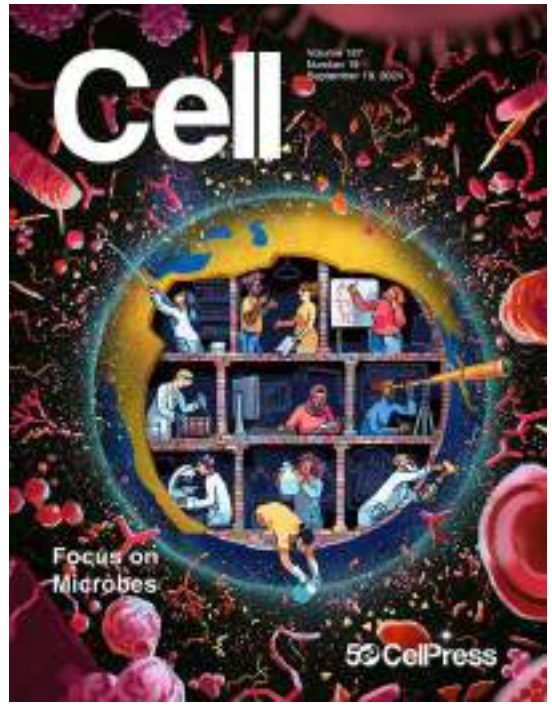
Conclusions

- Differences in the **diversity and composition** (relative abundance of taxa) of the gut microbiome singficantly impact the outcome of transplanted children.

- GM microbiota modulation is one of the most attractive scenario in the field of **HSCT**, **immunotherapy** and **cellular therapy**.

- A better understanding of the host-microbiota dialogue may contribute to pave the way for **precision medicine in pediatric hametological malignancies.**

Cell. Sept 19. 2024



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Thanks

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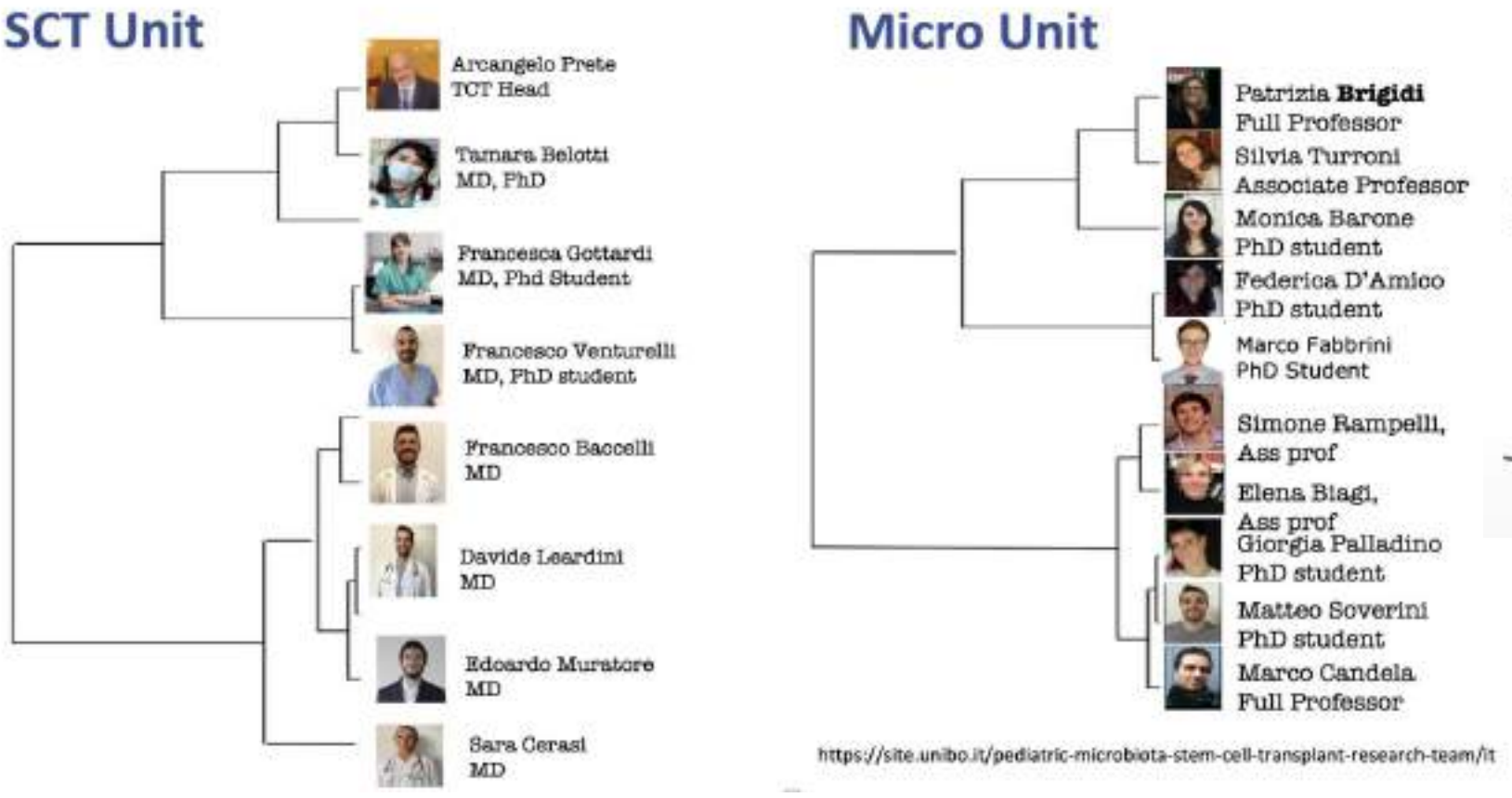
Prof. Lorenza Putignani

GRUPPO DI RICERCA SUL MICROBIOTA E TRAPIANTO DI CELLULE STAMINALI EMOPOIETICHE PEDIATRICO



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Dr. Ellie Margolis
Dr. Marygrace Duggar

Zeller Lab

Dr. Georg Zeller