Virome et Transplantation

SFGM-TC Scientific Day - *October 20, 2022*
« The gut microbiota »

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Human Immunology
Pathophysiology
Immunotherapy

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Hématologie I Immunologie I Oncologie

Disclosure Statement

No conflict of interest with this presentation

Virome

Bacteria

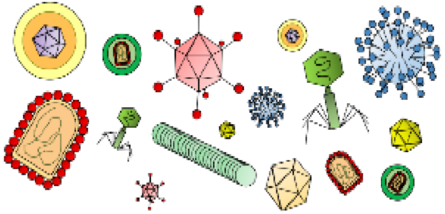
Fungi

Archaea

Eucaryotic Viruses

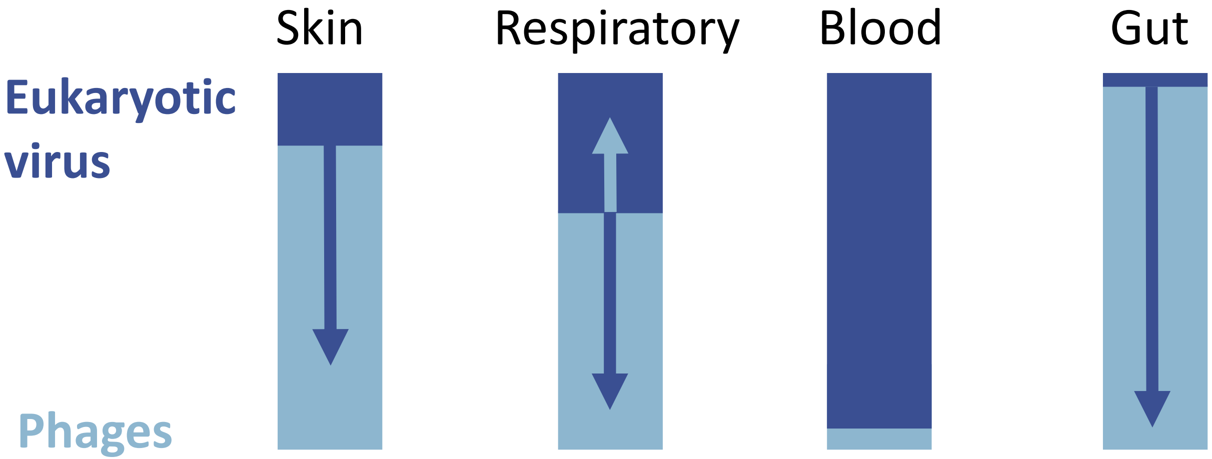
Bacteriophages

The virome



- Resident viruses (chronic infections)
- Exogenous viruses (transient infections)
- Endogenous retroviral elements
- Unclassified sequences
- **Pathogenic viruses ?**
- **Commensal viruses ?**

Virome composition



Skin Respiratory Blood Gut

Eukaryotic virus

Phages

Gut viromes are unique to individuals *Reyes A et al. 2010*

Healthy gut virome stable and dominated by phages

Variation between subjects according to the diet *Minot S et al. 2013*

↑Eukaryotic portion according to disease status

Diarrhea	*Phan TG et al. 2012*
Flacid paralysis	*Victoria J G et al. 2009*

10^{15} phages in the human intestine

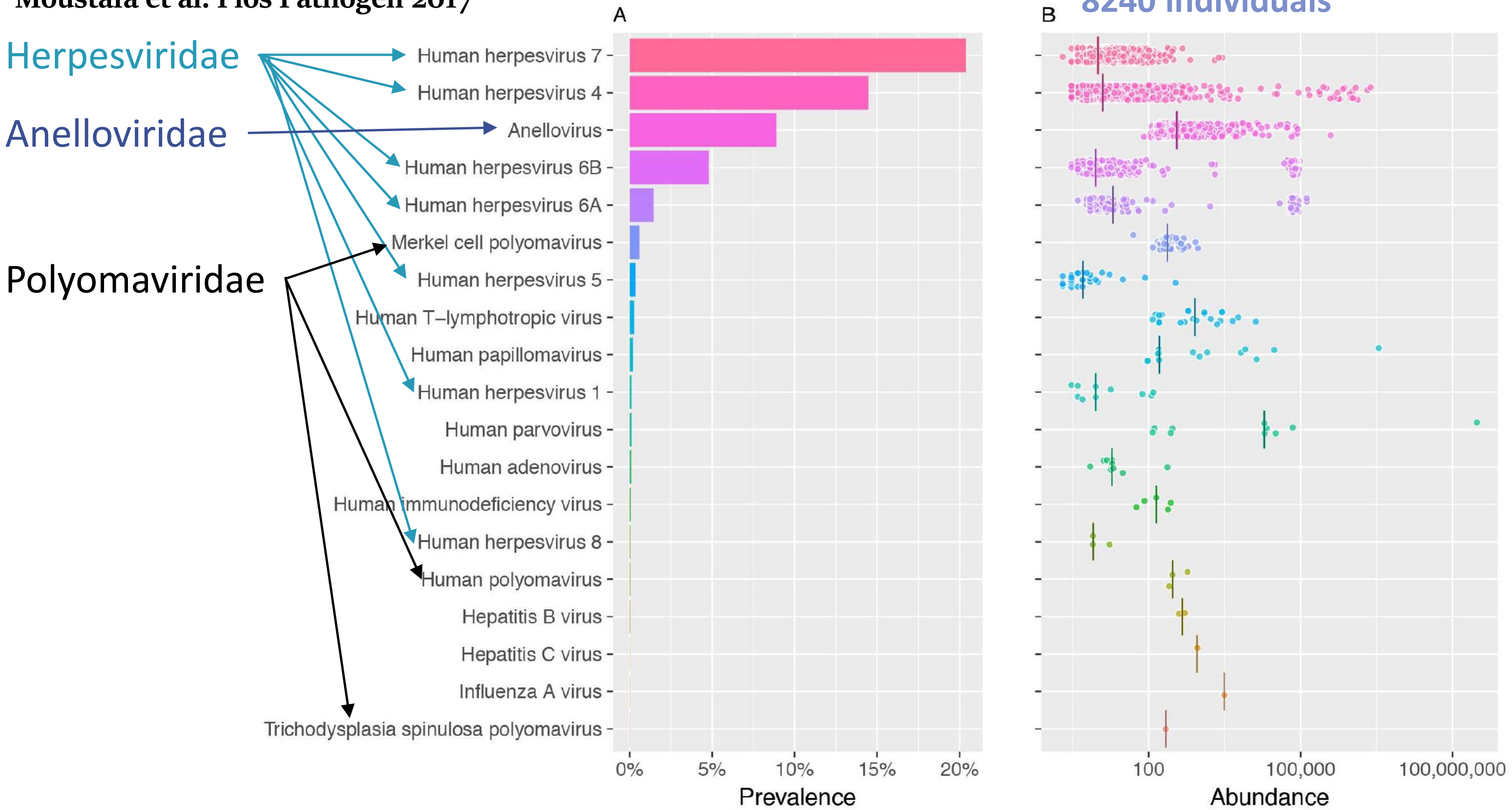
dsDNA Caudovirales & ssDNA *Microviridae*

1 g of stool →**10^{8} – 10^{9}** viruses

99% phages
1% animal/plant viruses

Normal blood Virome

Moustafa et al. Plos Pathogen 2017



8240 individuals

Herpesviridae

Anelloviridae

Polyomaviridae

Limits of detection of viruses

No « 16s » viral gene

Virus genome sizes

< $10^{e}2$ – $10^{e}4$ / bacteria

< $10^{e}4$ – $10^{e}6$ / eucaryotes

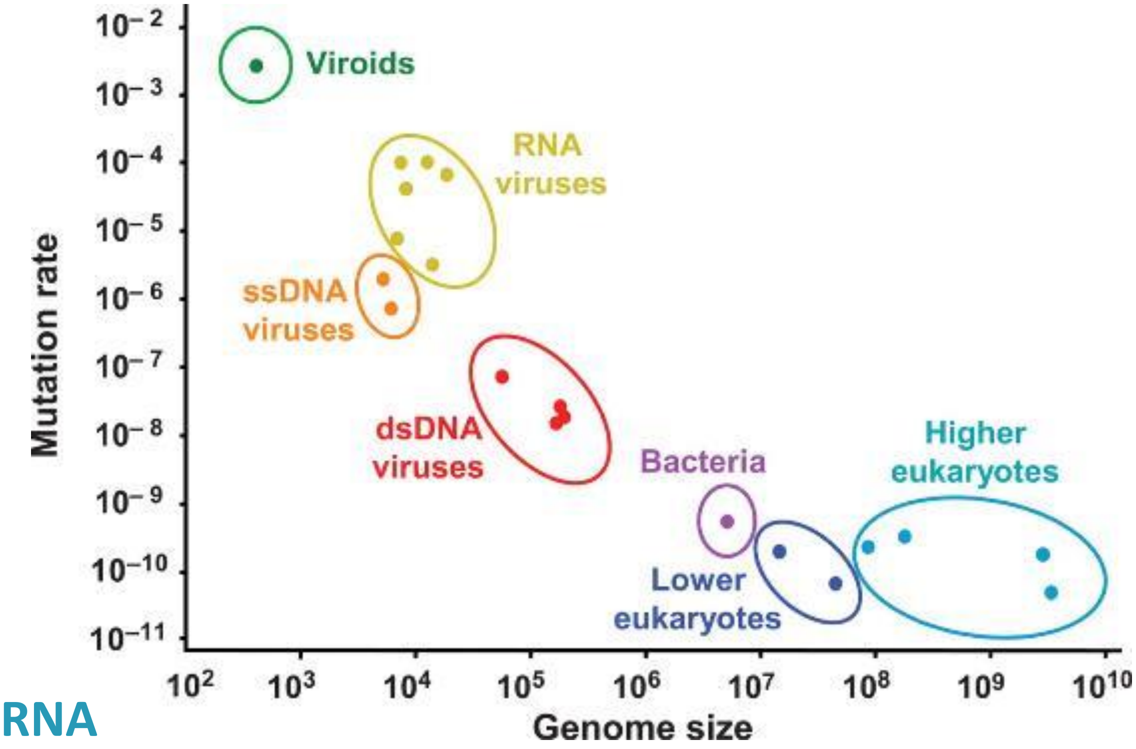
Human cell depletion

Bacteria depletion

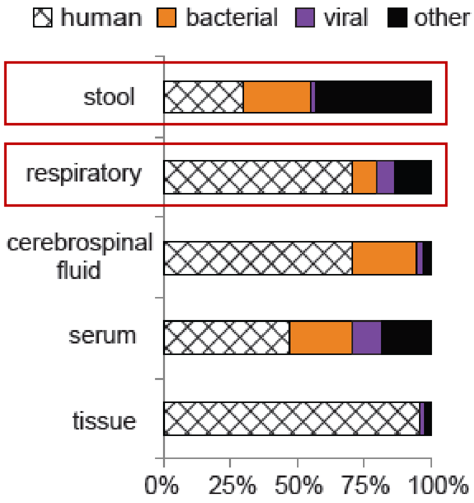
VLP enrichment

↗ reads

Amplification methods DNA & RNA

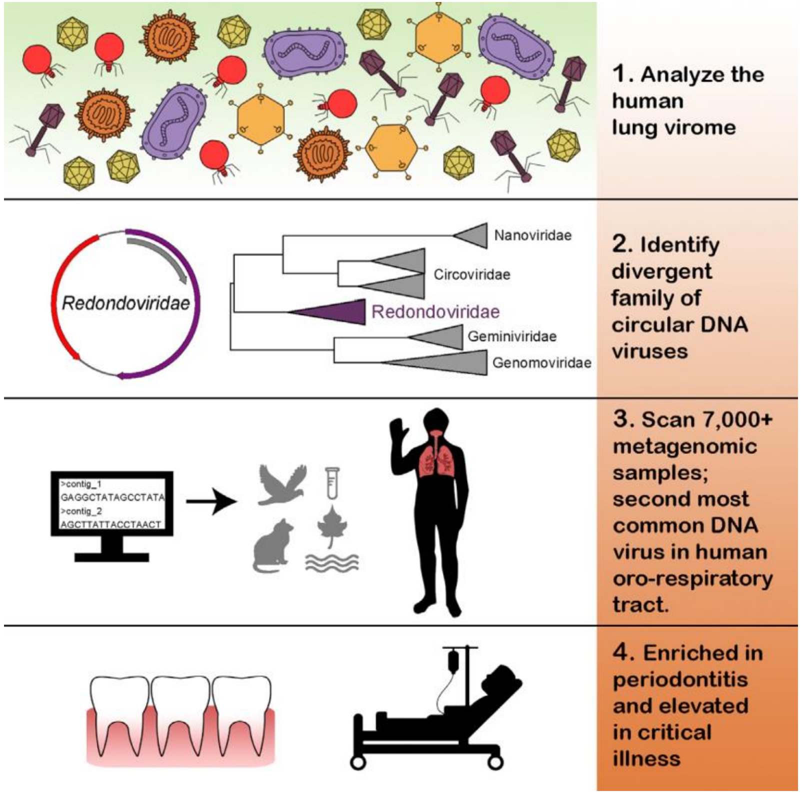


Limits of detection – Viral datbases



Abbas et al., 2019, Cell Host & Microbe

Redondoviridae, a Family of Small, Circular DNA Viruses of the Human Oro-Respiratory Tract Associated with Periodontitis and Critical Illness.



1. Analyze the human lung virome

2. Identify divergent family of circular DNA viruses

3. Scan 7,000+ metagenomic samples; second most common DNA virus in human oro-respiratory tract.

4. Enriched in periodontitis and elevated in critical illness

Limits of detection – Viral datbases

Cell

Resource

Expansion of the global RNA virome reveals diverse clades of bacteriophages

Neri et al., 2022, Cell 185, 1–15

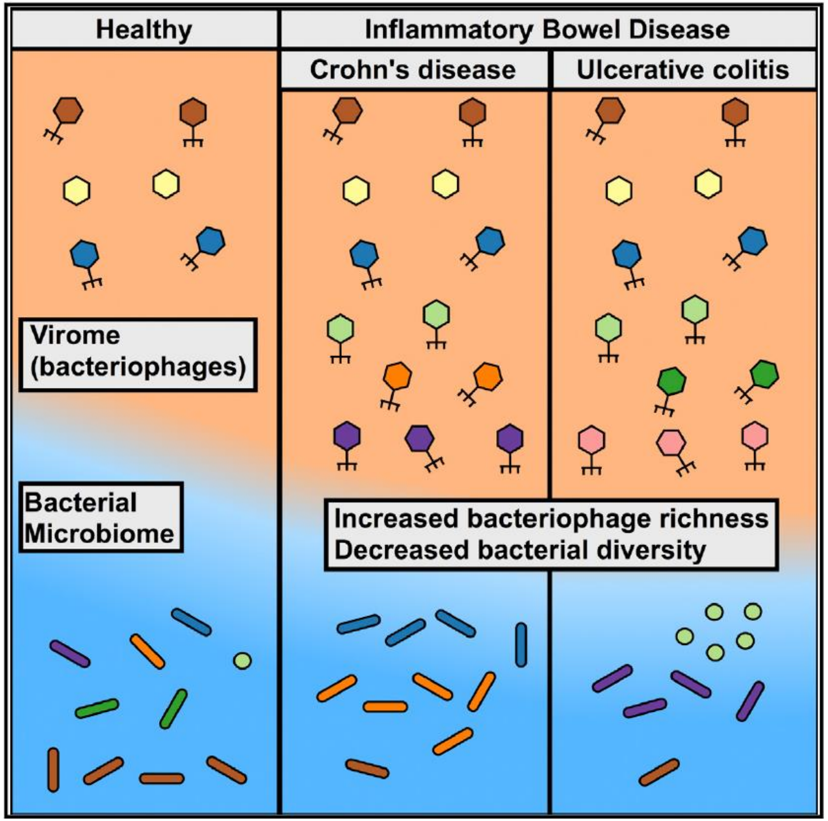
Analysis of >330,000 RNA-dependent RNA polymerases (RdRPs) shows that this expansion corresponds to **a 5-fold increase of the known RNA virus diversity**.

The dramatically expanded phylum **Lenarviricota**, consisting of bacterial and related eukaryotic viruses, now accounts for **a third of the RNA virome**.

Enteric Viruses and Colitis

Cell

Disease-Specific Alterations in the Enteric Virome in Inflammatory Bowel Disease



Norman et al., 2015, Cell *160*, 447–460

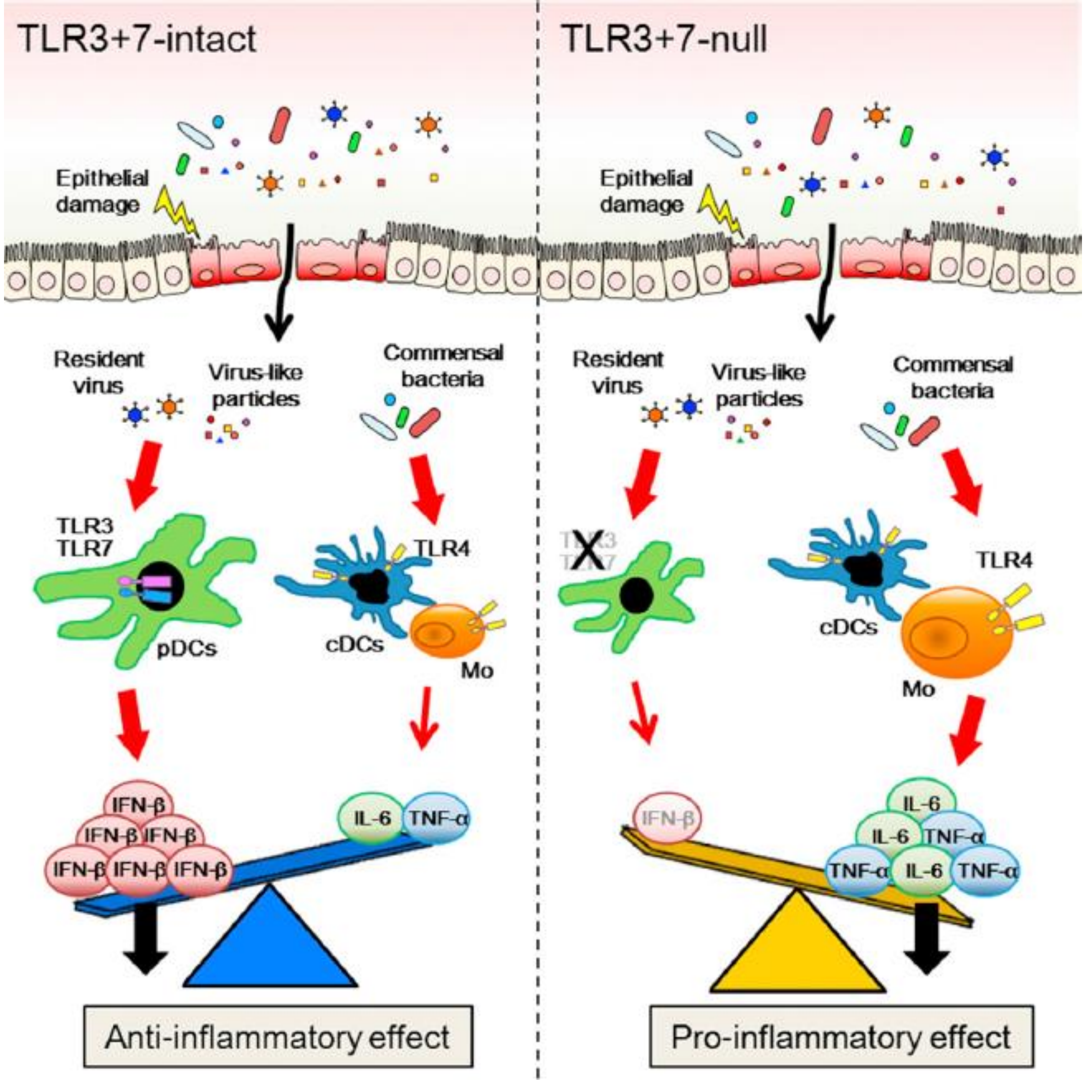
32000 reads / sample

- **The enteric virome richness increases in Crohn’s disease and ulcerative colitis**
- Decreases in bacterial diversity and richness in IBD do not explain virome changes
- **Virome changes in Crohn’s disease and ulcerative colitis are disease specific**

Enteric Viruses Ameliorate Gut Inflammation via Toll-like Receptor 3 and Toll-like Receptor 7-Mediated Interferon-β Production

Jin-Young Yang et al

Illumina
900,000 reads for DNA
690,000 reads for RNA



Enteric Viruses and Colitis

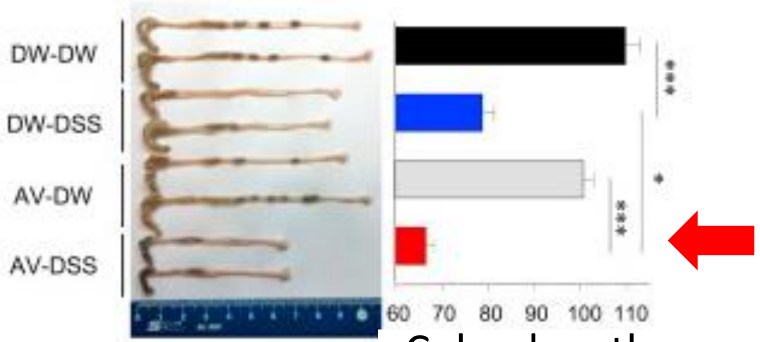
Jin-Young Yang et al. Immunity 2016

BALB/c wild-type (WT) mice +/- **antiviral (AV) cocktail** (ribavirin, lamivudine, acyclovir) 10 days + dextran sodium sulfate DSS treatment

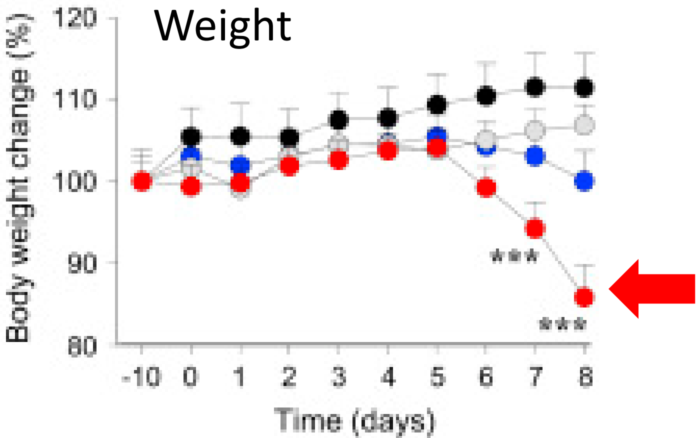
- Pre-treatment with an antiviral cocktail results in severe colitis
- Treatment with TLR3+7 agonists and inactivated rotavirus ameliorates colitis
- TLR3 -/- TLR7 -/- mice are more susceptible to colitis

Gut viruses → TLR3 or TLR7 ↗ IFN-beta

↘ inflammation



Colon length

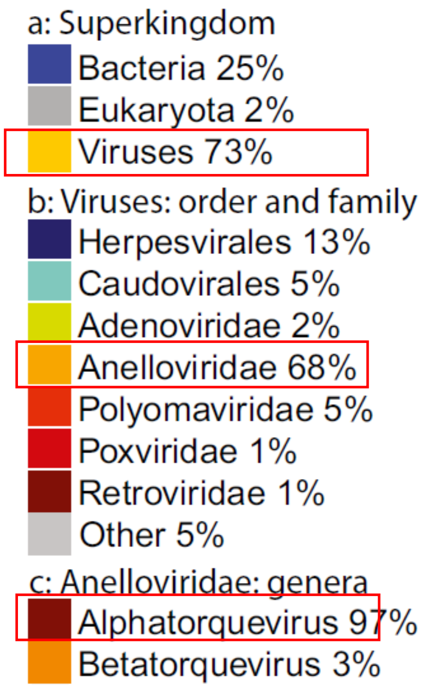


Virome et Transplantation

Heart and lung transplant

Blood virome – De Vlaminck et al, 2013. Cell

96 Heart/Lung transplant pt – 656 samples



a: Superkingdom
- 🟦 Bacteria 25%
- ⬜ Eukaryota 2%
- 🟨 Viruses 73%

b: Viruses: order and family
- 🟦 Herpesvirales 13%
- 🟩 Caudovirales 5%
- 🟨 Adenoviridae 2%
- 🟧 Anelloviridae 68%
- 🟥 Polyomaviridae 5%
- 🟥 Poxviridae 1%
- 🟫 Retroviridae 1%
- ⬜ Other 5%

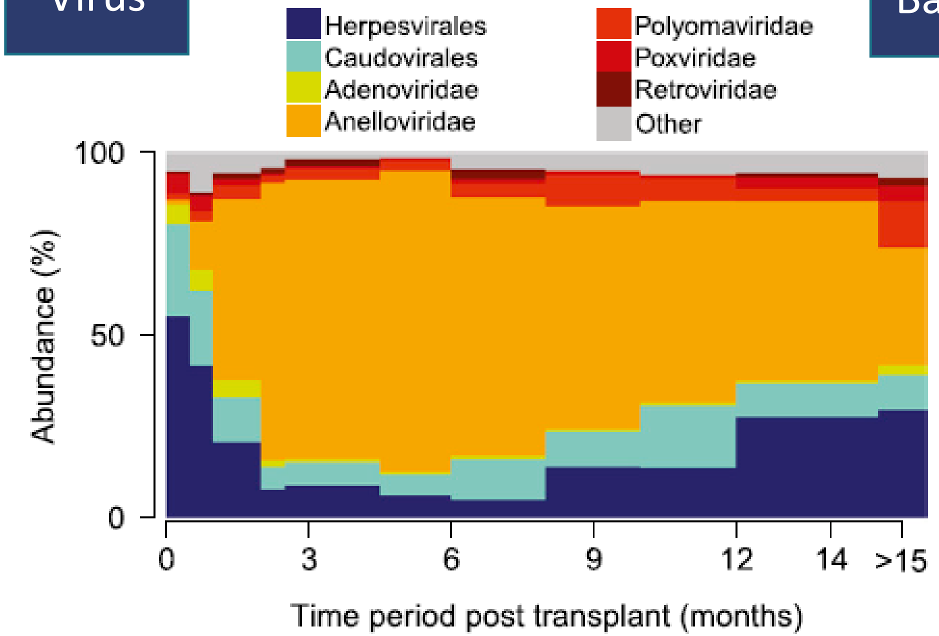
c: Anelloviridae: genera
- 🟫 Alphatorquevirus 97%
- 🟧 Betatorquevirus 3%

Heart and lung transplant

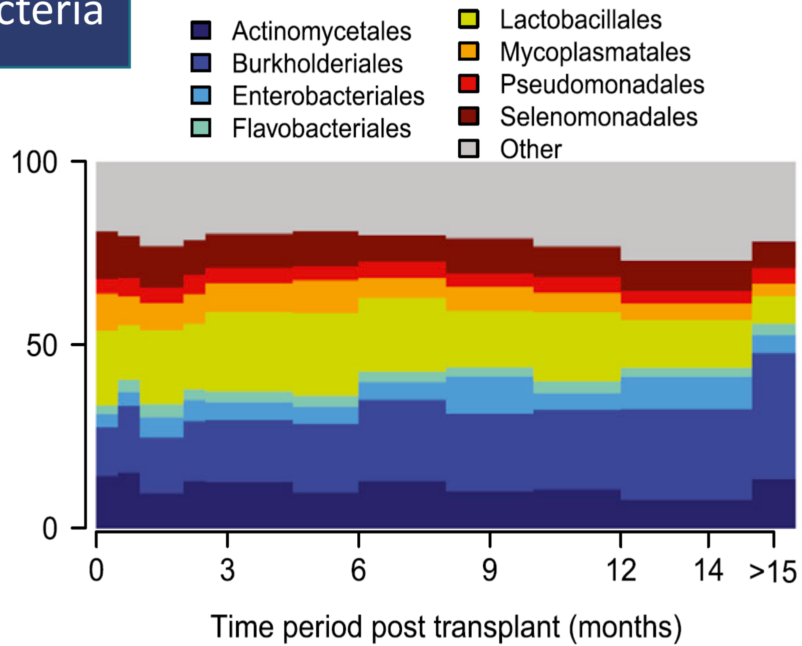
Temporal change of the virome De Vlaminck et al, 2013. Cell

No change of the bacterial microbiome over time

Virus

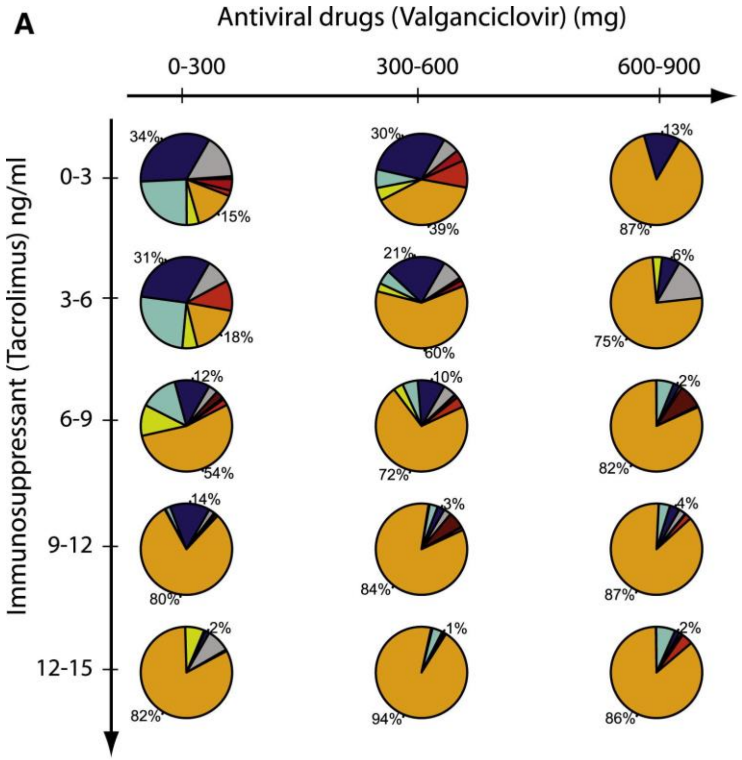


Bacteria



Impact of antivirals & immunuosuppressants

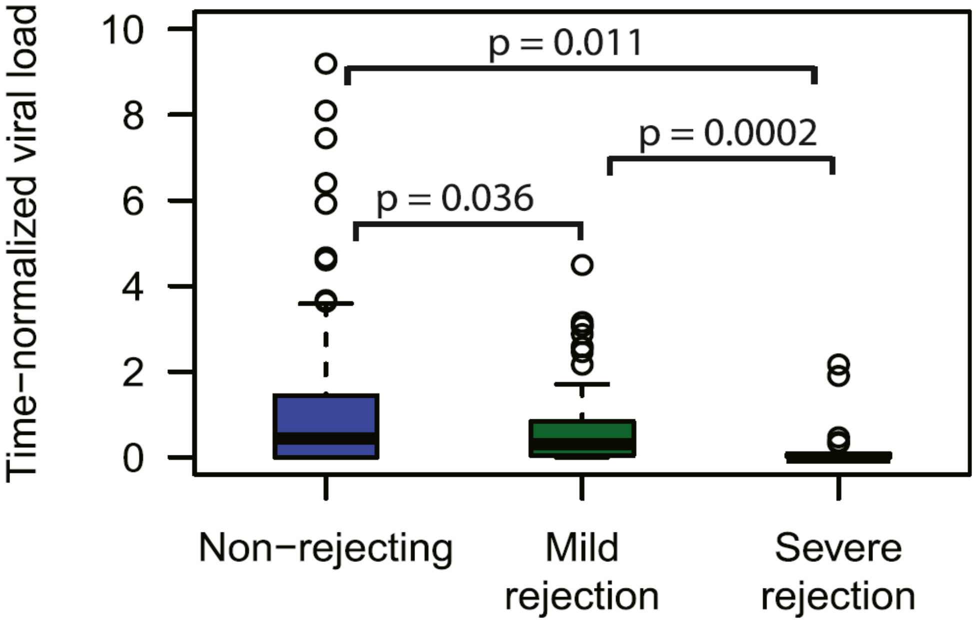
Tacrolimus-based antirejection protocol (47 patients and 380 observations)



Antivirals Immunosuppressants	Herpes Phages	Anellovirus
Low dose	↑	↓
High dose	↓	↑

Correlation with graft rejection

↘ Anellovirus burden in patients with graft rejection



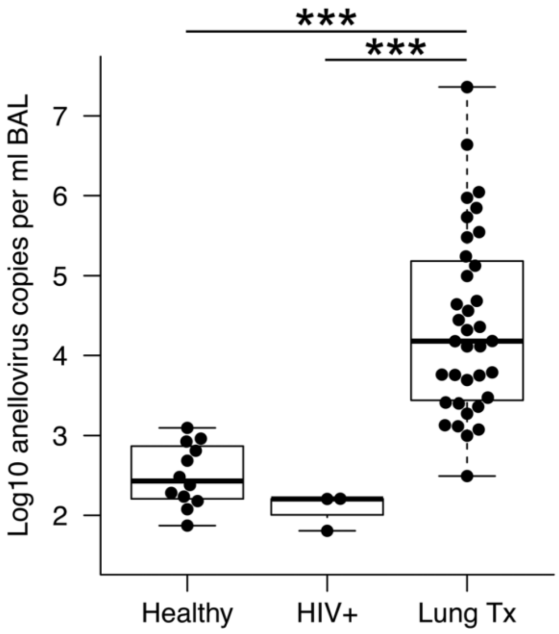
➢ Alternative marker to measure level of immunosupression

Lung Transplant Young, Am J Transplant. 2015

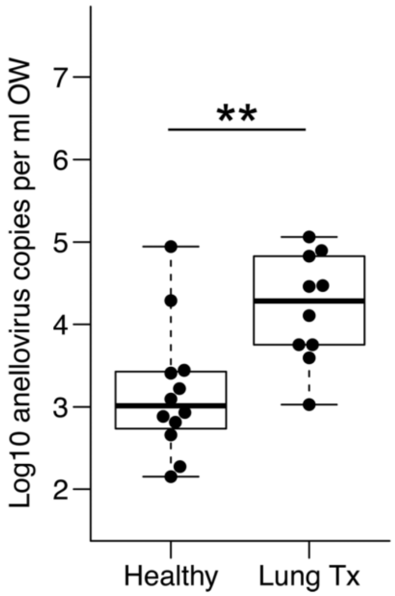
Anellovirus **56-fold more abundant** in **BAL** from lung Tx

Anellovirus loads correlated with dysbiotic bacterial communities

BAL



ORAL WASHES

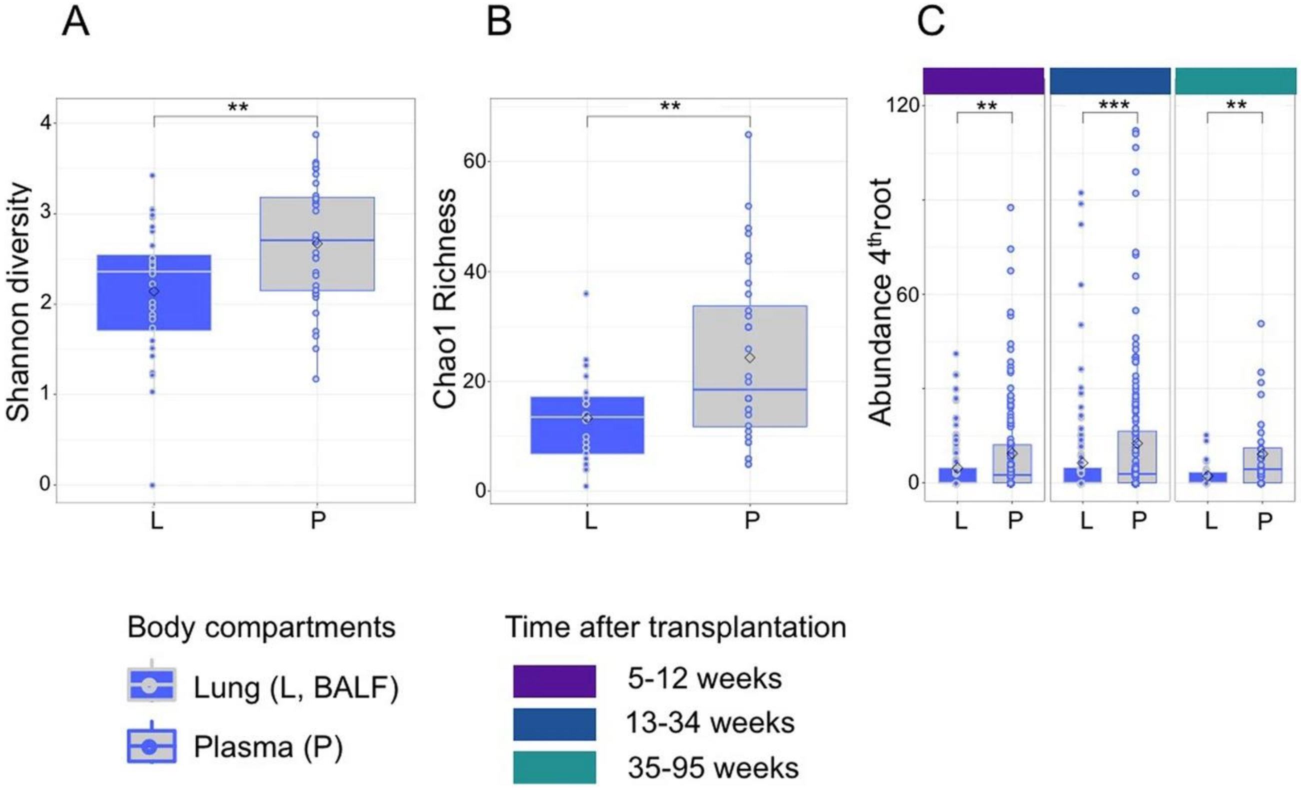


Virome in the Lungs: The Role of Anelloviruses in Childhood Respiratory Diseases. Giulia Dodi et al. Microorganisms. 2021.

Lung Transplant

Metagenomic sequencing reveals time, host, and body compartment-specific viral dynamics after lung transplantation. Widder et al. Microbiome 2022

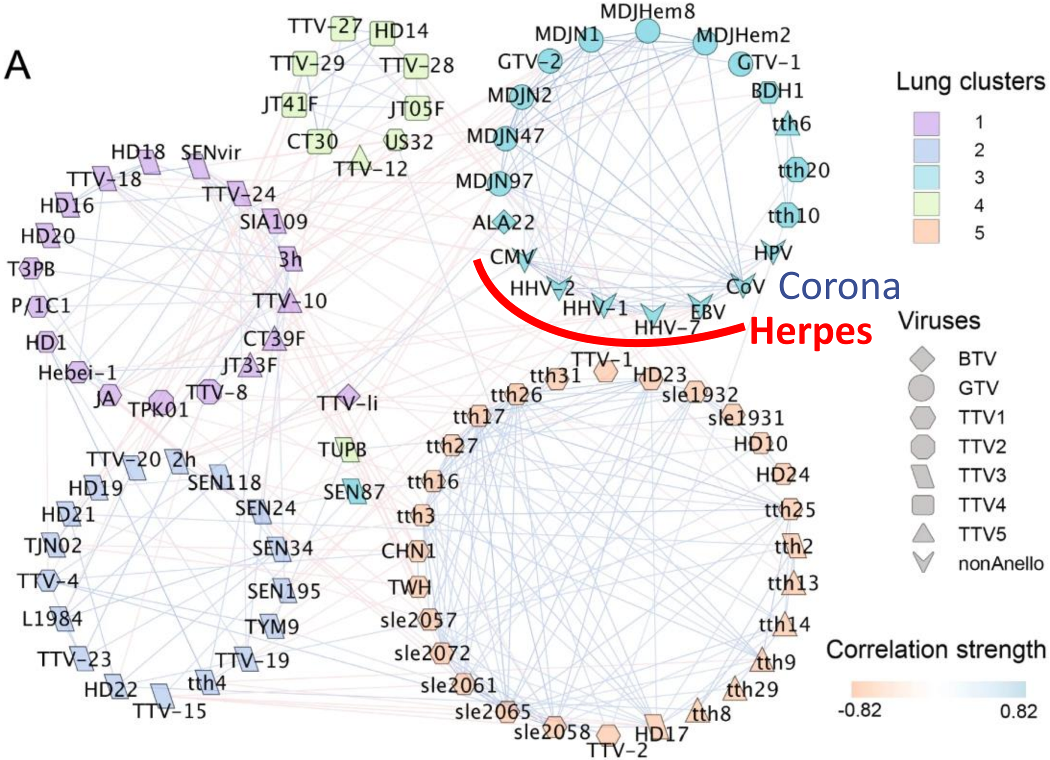
Higher taxonomic diversity in plasma samples compared to BALF

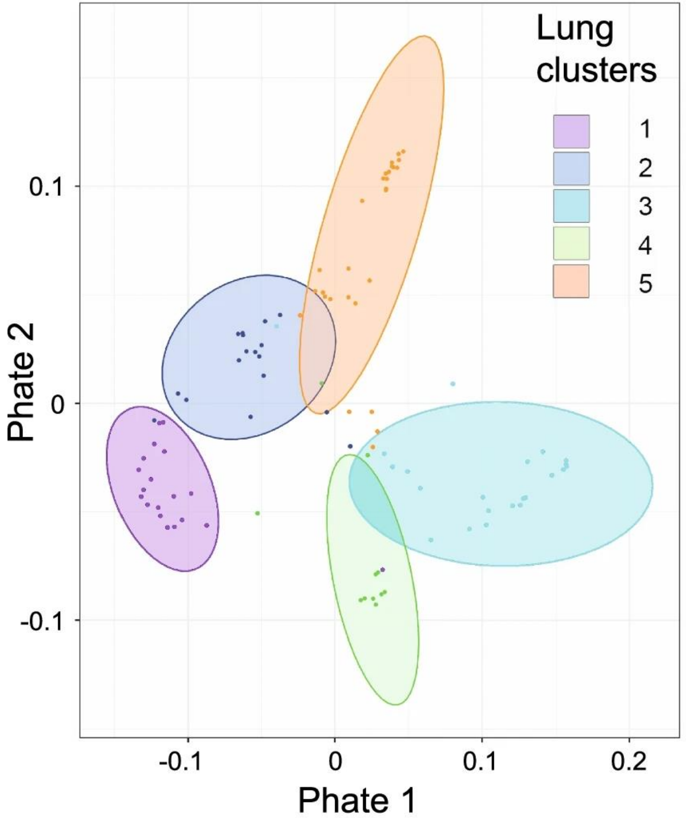


Lung Transplant

Widder et al. Microbiome 2022

Exclusive viral communities in BALF and plasma

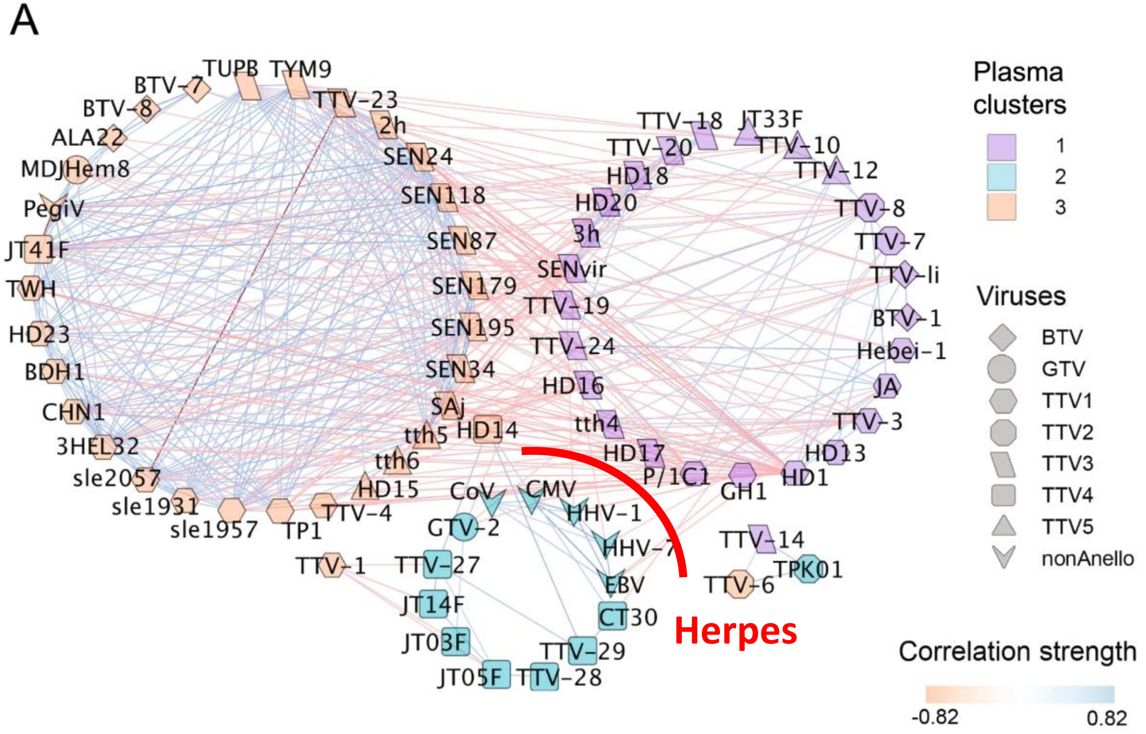




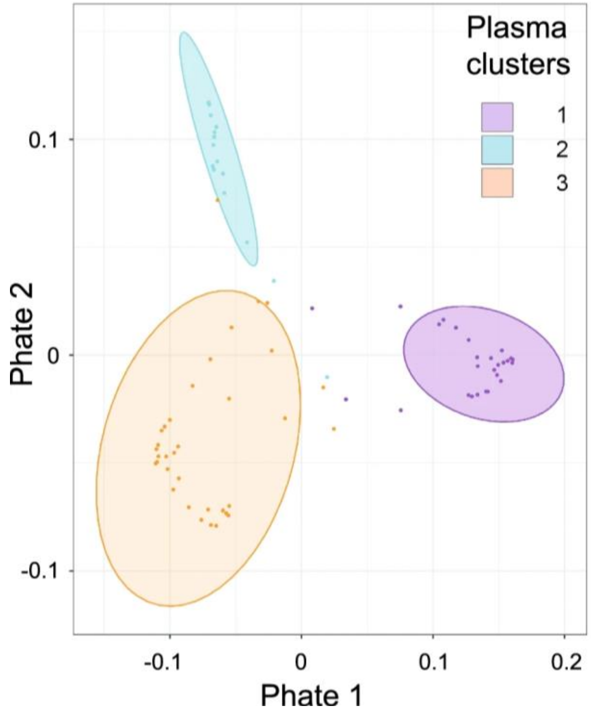
Lung Transplant

Widder et al. Microbiome 2022

Exclusive viral communities in BALF and plasma



Herpes

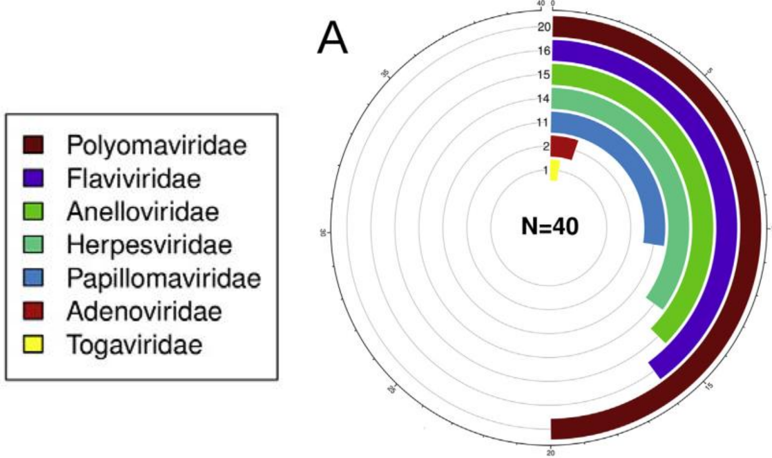


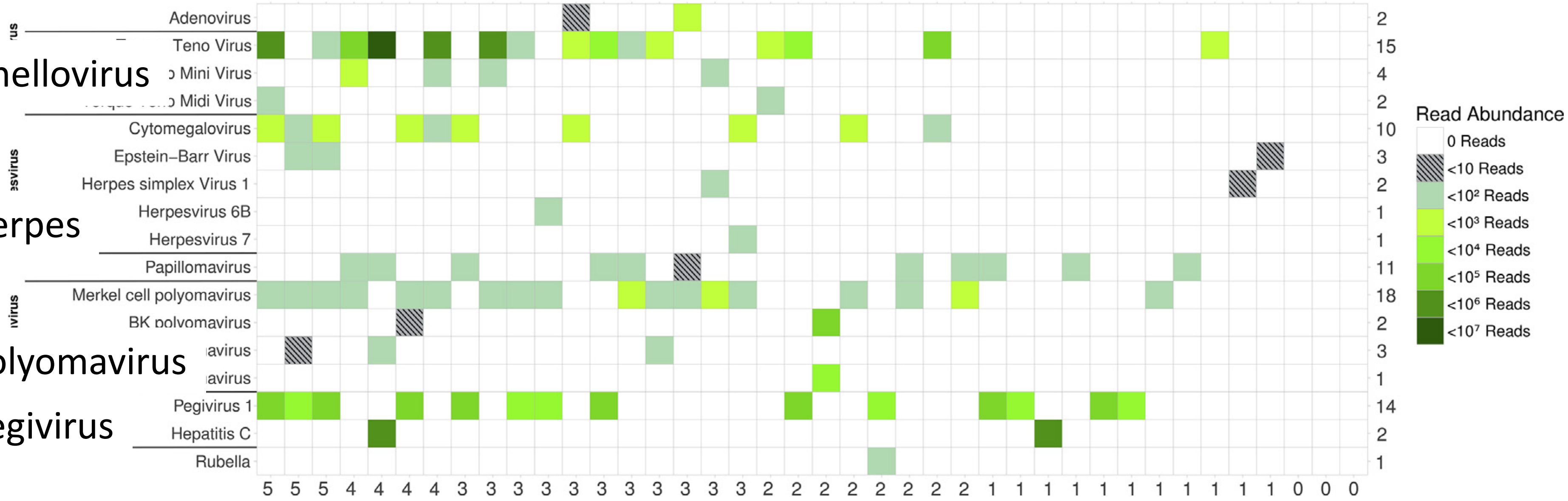
Recipients' underlying disease imprints long-term plasma virome structure

Blood virome in hematopoietic stem cell transplantation

Vu DL et al. Clin Microb Infect 2019

1 month after transplantation





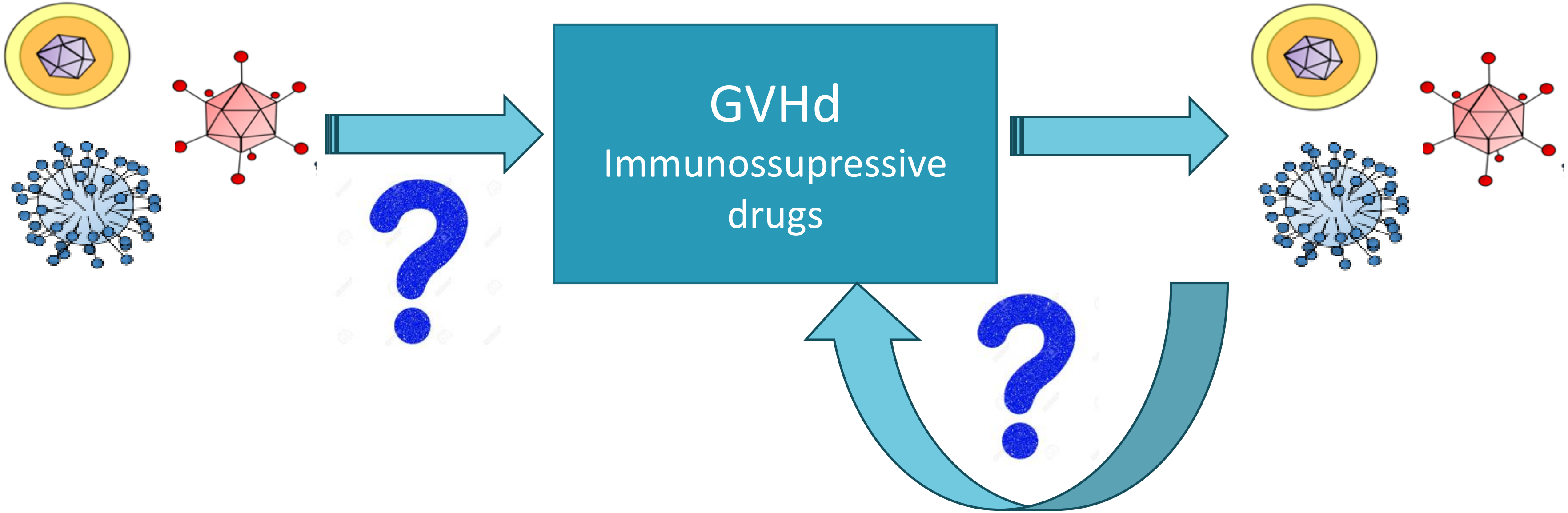
Anellovirus

Herpes

Polyomavirus

Pegivirus

Virus and Graft versus host disease



GVHd

Immunossupressive drugs

Viruses & GvHD

Herpesviruses

- CMV (Blood) CMV ➚ 2X GVHD risk - *Cantoni et al, 2010*
- HHV-6 (Blood) - *Pichereau et al., 2012, Zerr et al., 2012*
- HSV-1 (Skin – CD34+ PBMC) - *Akpek et al,.2013*
- EBV serostatus. D+/R+ HR=1.24 - *J Clin Oncol. 2016 34(19):2212-20*

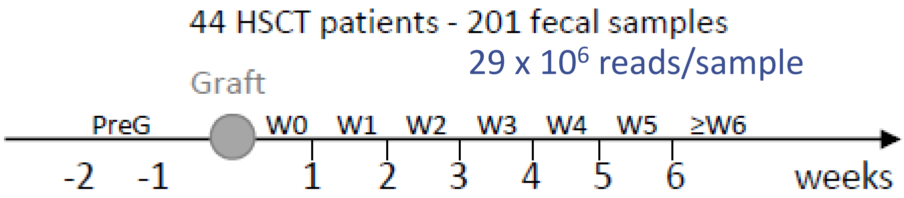
Adenoviruses

Mechanism ?

Virus mimicry → Up to 45% of virus specific T-cell clones crossreact against allogeneic HLA molecules (Amir AL, Blood. 2010)

Gut Virome in HSCT

The eukaryotic gut virome in HSCT : new clues in enteric GvHD. *Nat Med. 2017 Sep;23(9):1080-1085.*



44 HSCT patients - 201 fecal samples

29 x 10^6 reads/sample

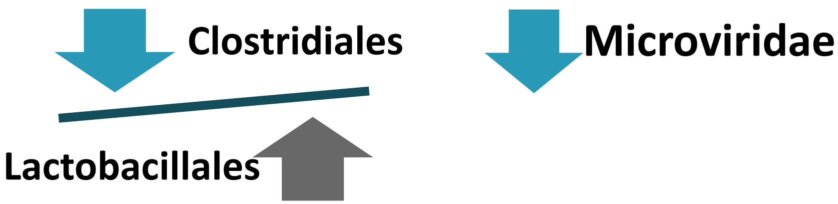
Median % of viral reads = **0.22%**

→ Eukaryotic viruses =**1.76% of viral reads**

IQR. 0.16–21.3%   Range. **0–99.9%**

Vertebrate viruses = 86.9% - Plant viruses = 12.1%

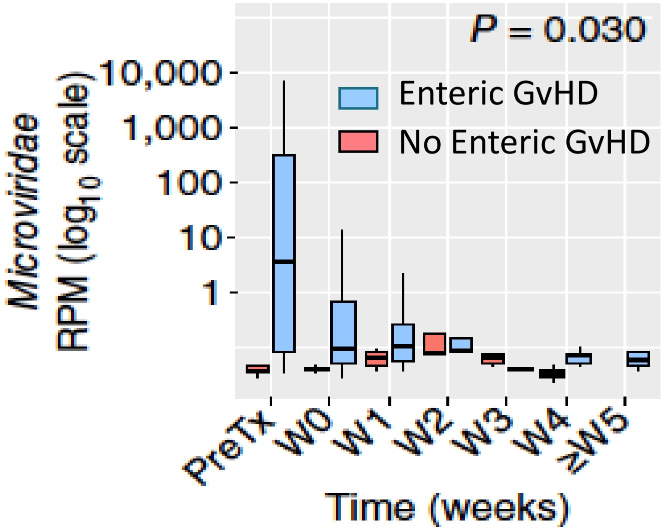
Gut Microbiota   Gut Phages



Clostridiales

Lactobacillales

Microviridae



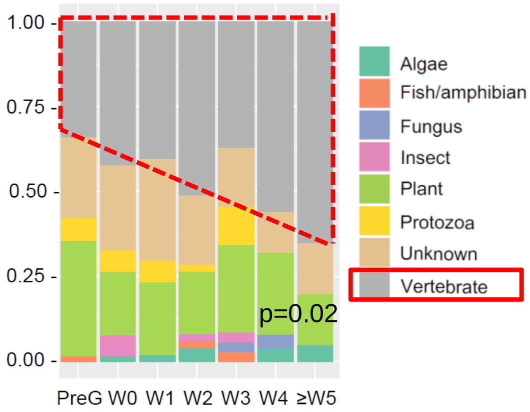
Gut Virome in HSCT

The eukaryotic gut virome in HSCT : new clues in enteric GvHD. *Nat Med. 2017 Sep;23(9):1080-1085.*

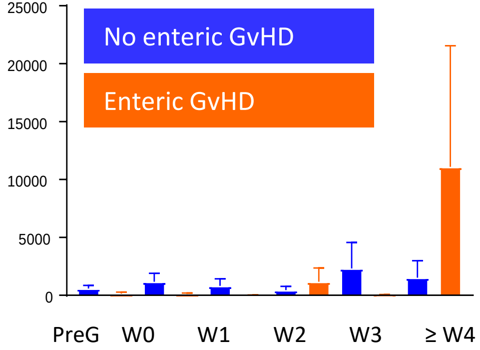
Gut Eukaryotic viruses

→ Increase of vertebrate viruses

→ Increased rates and loads of persistent viruses in patients with GvHD



Persistent viruses

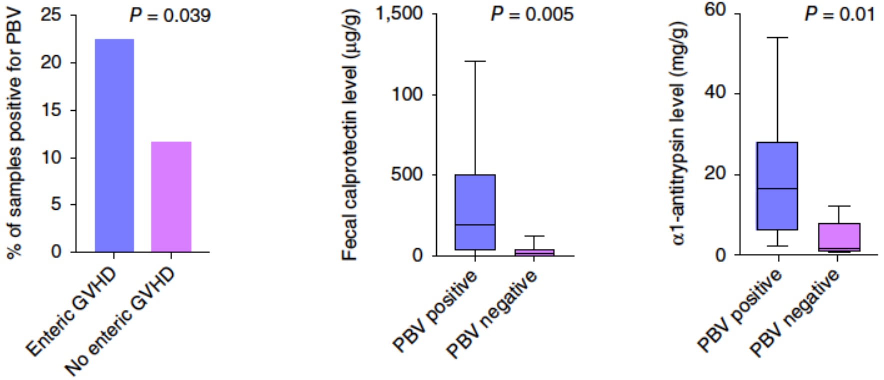


→ Detection of **Picobirnavirus** predictive of GvHD

Time dependent Cox model (HR:2,66)

→ Picobirnavirus associated with enteric inflammation

↗ Fecal calprotectin – alpha1 antitrypsin



Picobirnavirus – Eukaryotic virus or Phage

EUKARYOTIC ?

Detection in stool samples of AIDS patients

Detection in stool samples of many mammalian species

Picobirnavirus related to Partitivirus (yeast and

Duquerroy S et al. EMBO J. 2009.

PHAGES ?

Presence of prokaryotic ribosomal bindi

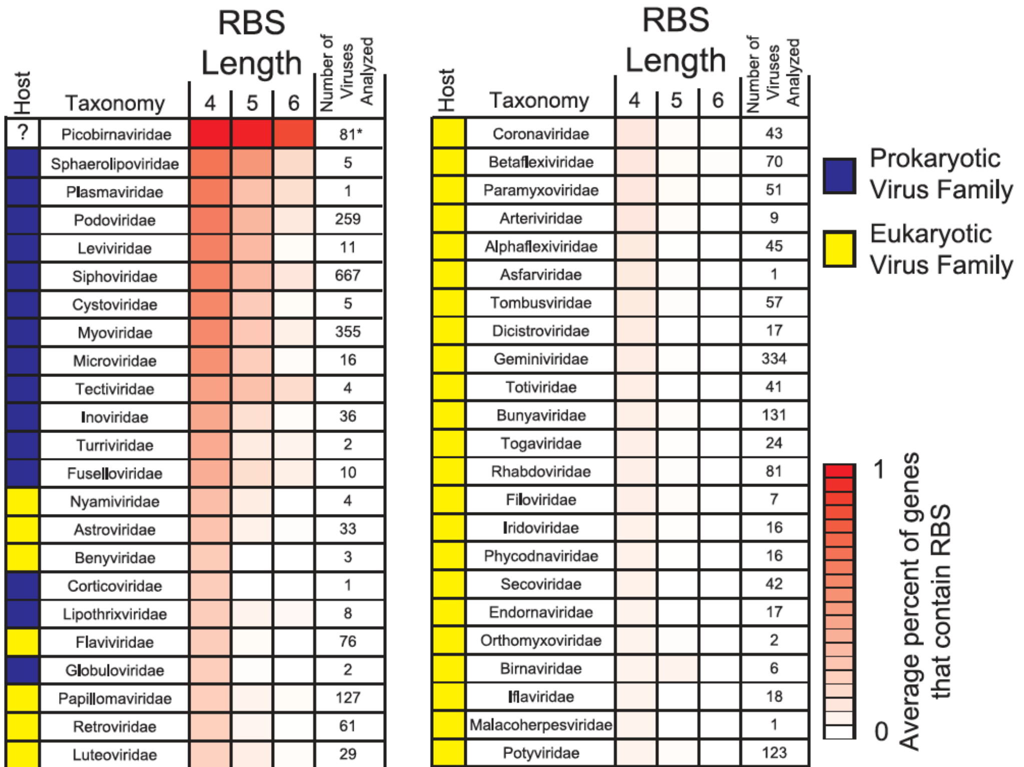
- Krishnamurthy SR, Wang D. Virology. 2018
- Boros Á et al. Virology. 2018

Ribosomal binding sites -10 -15 nt upstrean

4 mers (AGGA, GGAG, GAGG) - 5 mers (AGGAG, GG

Identification of CRISPR spacer matches picobirnaviruses and partitiviruses, prev hosts.

- Neri et al., 2022, Cell 185, 1–15



Host	Taxonomy	RBS Length 4	5	6	Number of Viruses Analyzed
?	Picobirnaviridae				81*
🟦	Sphaerolipoviridae				5
🟦	Plasmaviridae				1
🟦	Podoviridae				259
🟦	Leviviridae				11
🟦	Siphoviridae				667
🟦	Cystoviridae				5
🟦	Myoviridae				355
🟦	Microviridae				16
🟦	Tectiviridae				4
🟦	Inoviridae				36
🟦	Turriviridae				2
🟦	Fuselloviridae				10
🟨	Nyamiviridae				4
🟨	Astroviridae				33
🟨	Benyviridae				3
🟦	Corticoviridae				1
🟦	Lipothrixviridae				8
🟨	Flaviviridae				76
🟦	Globuloviridae				2
🟨	Papillomaviridae				127
🟨	Retroviridae				61
🟨	Luteoviridae				29

Host	Taxonomy	RBS Length 4	5	6	Number of Viruses Analyzed
🟨	Coronaviridae				43
🟨	Betaflexiviridae				70
🟨	Paramyxoviridae				51
🟨	Arteriviridae				9
🟨	Alphaflexiviridae				45
🟨	Asfarviridae				1
🟨	Tombusviridae				57
🟨	Dicistroviridae				17
🟨	Geminiviridae				334
🟨	Totiviridae				41
🟨	Bunyaviridae				131
🟨	Togaviridae				24
🟨	Rhabdoviridae				81
🟨	Filoviridae				7
🟨	Iridoviridae				16
🟨	Phycodnaviridae				16
🟨	Secoviridae				42
🟨	Endornaviridae				17
🟨	Orthomyxoviridae				2
🟨	Birnaviridae				6
🟨	Iflaviridae				18
🟨	Malacoherpesviridae				1
🟨	Potyviridae				123

🟦 Prokaryotic Virus Family

🟨 Eukaryotic Virus Family

Gut Virome in HSCT

Different gut virome dynamics in patients with or without GvHD

Decrease in Microviridae – similar to phage dynamics in IBD

Expansion of vertebrate viruses after HSCT

- ➢ Increase of persistent DNA viruses in GvHD
- ➢ Stability of persistent DNA viruses in patients without GvHD
- ➢ Picobirnavirus detection marker of GvHD

Virome & Transplantation

- Clusters according to underlying disease
- Compartmentation and temporal dynamics of viromes
- Viruses /virome predictive of graft versus host disease
- Phages / RNA virome and interactions with microbiota and eukaryotic viruses poorly understood

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Acknowledgments

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