UNDERSTANDING THE EFFECT OF DRUGS ON MICROBIOME DYNAMICS USING MATHEMATICAL MODELS

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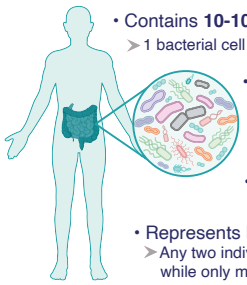
1 The gut microbiome



- Contains **10-100 trillion cells**
  - 1 bacterial cell for every human cell[1]
- Mediates **immune regulation**
- Releases crucial **vitamins, amino acids** & **neurotransmitters**
- Contains **> 150x more genes** than in human genome
- Represents large **inter-individual variability**
  - Any two individuals share >90% of human DNA, while only max. 10% of bacterial DNA
- **Mediates** therapeutic **drug composition** & effects[3]

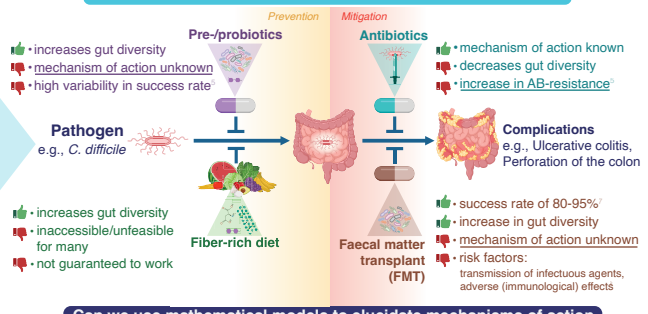
Gut pathogenicity in the UK

- **17 million cases** of gastrointestinal infections each year[4]

1 in 5 people affected annually

- Important agent of disease: ***Clostridioides difficile* infection (CDI)**
  - CDI causes **20-30%** of antibiotics-associated **diarrhoea**[5]
  - Common **after antibiotics course**
  - **Burden on UK Healthcare**: >£1713-£5126 for each hospitalised CDI case[6]

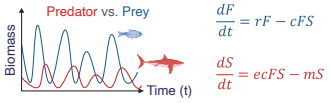
Current Interventions & Challenges



Can we use mathematical models to elucidate mechanisms of action for probiotics/FMT-based interventions?

2 Mathematical models - how do they work?

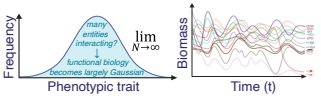
- Adapting classic ecological models used to describe & predict population dynamics e.g., Lotka-Volterra predator-prey equations:



$$\frac{dF}{dt} = rF - cFS$$

$$\frac{dS}{dt} = ecFS - mS$$

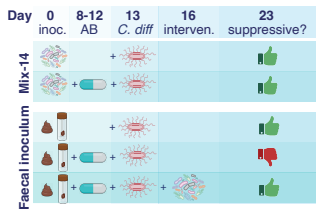
- Combine with concepts of statistical physics



$$\frac{dB_i}{dt} = \left(r_i B_i \left(1 - \frac{\sum_{i=1}^{N+1} \alpha_{i,j} B_i}{K_i}\right)\right)$$

Arbitrarily many taxa can now **be modelled**: required for microbiome

3 Mystery: Mix of 14 gut bacteria suppresses *C. difficile* after antibiotics



***In vitro* observations:**

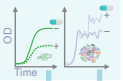
- Mix of 14 known gut bugs (Mix-14) **recovers suppressiveness** of faecal community after AB
- Yet, any Mix-14 member by itself not suppressive enough:
  - implies **community-dependency**

4 Using models to infer community interactions driving suppression

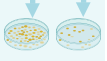
Data used for model input

Bacterial relative abundance (16S rDNA)
- Clustering of bacteria

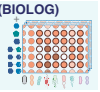
Optical Density (OD)
- Growth kinetics (with and w/o anti-biotics)



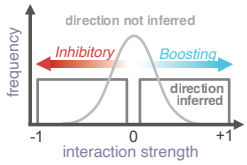
***C. difficile* load (CFUs)**
- Suppressive capacities (w and w/o antibiotics)

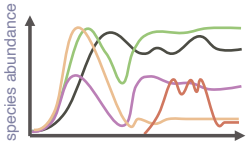


Carbon utilisation assay (BIOLOG)
- Similarity in substrate use

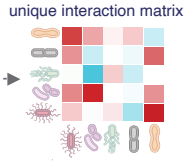


Simulation steps





unique interaction matrix



equations & integration designed to 'match' experimental timeline

>1.5 million configurations tested

α-diversity & suppressiveness within range observed *in vitro*?

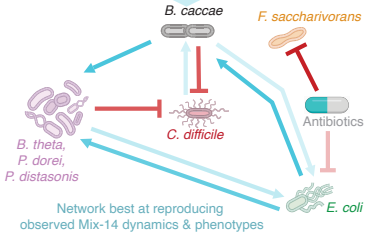
5 Mathematical model helps discover novel microbial and molecular signatures of *C. difficile* suppression

$$\frac{dB_i}{dt} = \left(\left(r_i + \sum_{j=1}^{N} \alpha_{i,j} B_j\right) B_i \left(1 - \frac{\sum_{i=1}^{N+1} B_i}{K}\right)\right) x - \phi B_i \quad \text{for } i \neq C.difficile$$

$$x = \begin{cases} 1 \text{ if } \phi = 0 \\ 1 - \gamma_1 \text{ otherwise} \end{cases}$$

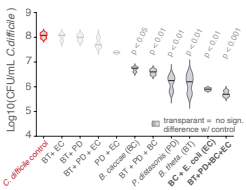
$$\frac{dB_i}{dt} = \left(r_i + \sum_{j=1}^{N} \alpha_{i,j} B_j\right) B_i \left(1 - \frac{B_i}{K_i}\right) - \phi B_i \quad \text{for } i = C.difficile$$

Following > million simulations, the following network most probable to reproduce observed composition & traits of Mix-14



Network best at reproducing observed Mix-14 dynamics & phenotypes

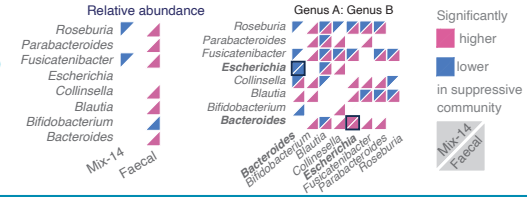
Model output validation 1



Ratio between *Bacteroides & Escherichia* key in *C. difficile* suppression

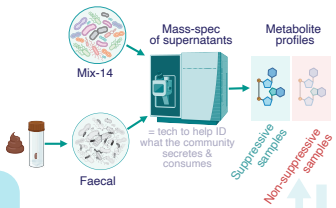
Both in Mix-14 & Faecal communities

Model output validation 2

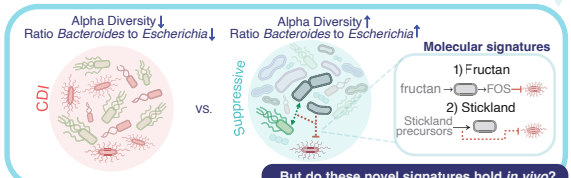


What **molecular mechanisms** drive observed **suppression**

& link to compositional signatures?



Use compositional signatures to guide 'hunt' for **relevant metabolites**

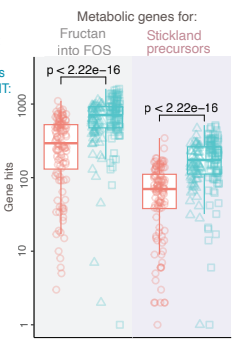


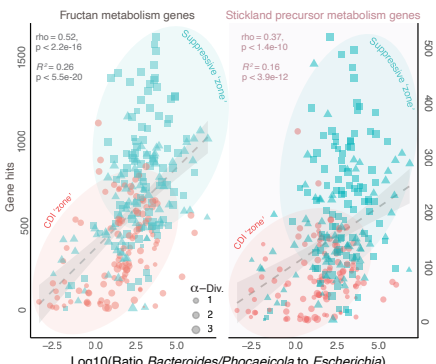
But do these novel signatures hold *in vivo*?

6 Novel molecular & compositional signatures of *C. diff* suppression validated across patient metagenomes

CDI-Suppressive:
- Healthy: N = 251, 10 cohorts
- Post-FMT: N = 222, 6 cohorts

CDI: N = 216, 8 cohorts





7 Conclusions & Potential applications

- Effectiveness probiotic mixes or FMT in **suppressing CDI** depends on **microbial ratios**; e.g., ratio of *Bacteroides* to *Escherichia*
  - Explains large variability in success rates of probiotic mixes
- Our **model-guided** framework helped discover **novel biomarkers** for **personalised probiotic therapy**
- We recommend **future work** to investigate microbial ratios to find biomarkers **otherwise invisible** to unguided statistical analysis of multi-omics data

Next steps: testing effectiveness probiotic mixes at different ratios in a controlled randomised trial

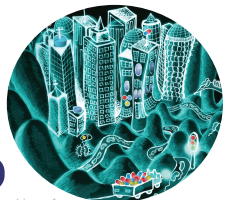


Image by Author: Gut Microbiome As a Metropolitan City: if we learn when and where to zoom in (or out), order appears

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[C] Currently: Stanford University

[1] Sender et al., (2016). Revised estimates for the number of human and bacteria cells in the body. *PLoS biology*, 14(8), e1002533.
[2] Zhu & Wang (2010). Human gut microbiome: the second genome of human body. *Protein & cell*, 1(8), 718-725.
[3] Klünemann et al. (2021). Bioaccumulation of therapeutic drugs by human gut bacteria. *Nature*, 597(7877), 533-538.
[4] Public Health England. Accessed February 2024. <https://www.gov.uk/government/collections/gastrointestinal-infections-guidance-data-and-analysis>
[5] UK Health Security Agency, CDI Guideline, July 2022.
[6] Robertson et al., (2020). Cost burden of Clostridioides difficile infection to the health service: a retrospective cohort study in Scotland. *Journal of Hospital Infection*, 106(3), 554-561.
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[8] Michalidis et al., (2021). Adverse events of faecal microbiota transplantation: A meta-analysis of high-quality studies. *Annals of Gastroenterology*, 34(6), 802.

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