

# top ten

in gastroenterologia

**14<sup>^</sup> EDIZIONE**

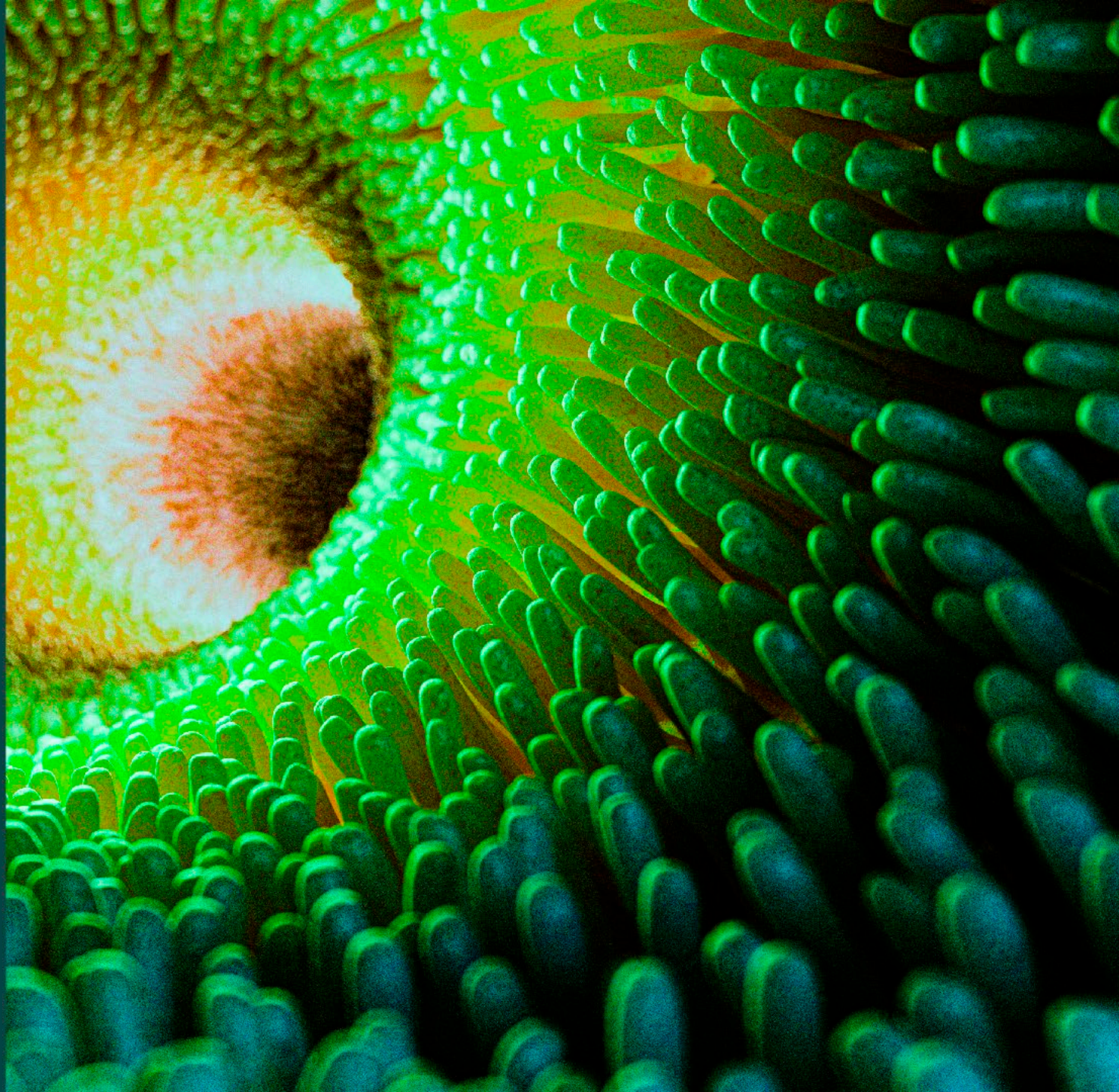
---

**24-25 NOVEMBRE 2023**

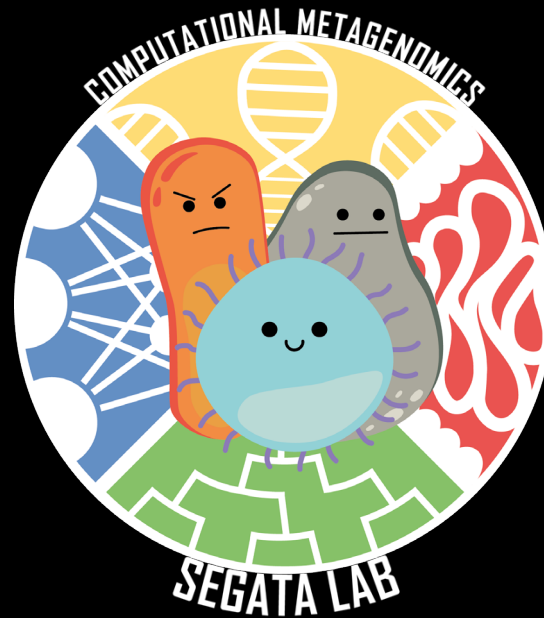
---

BERGAMO

HOTEL EXCELSIOR SAN MARCO  
Piazza della Repubblica, 6



# Risposta postprandiale al pasto e nutrizione di precisione



 @cibiocm

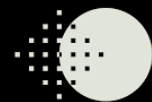
 @nsegata

Nicola Segata

November 2022



UNIVERSITY  
OF TRENTO

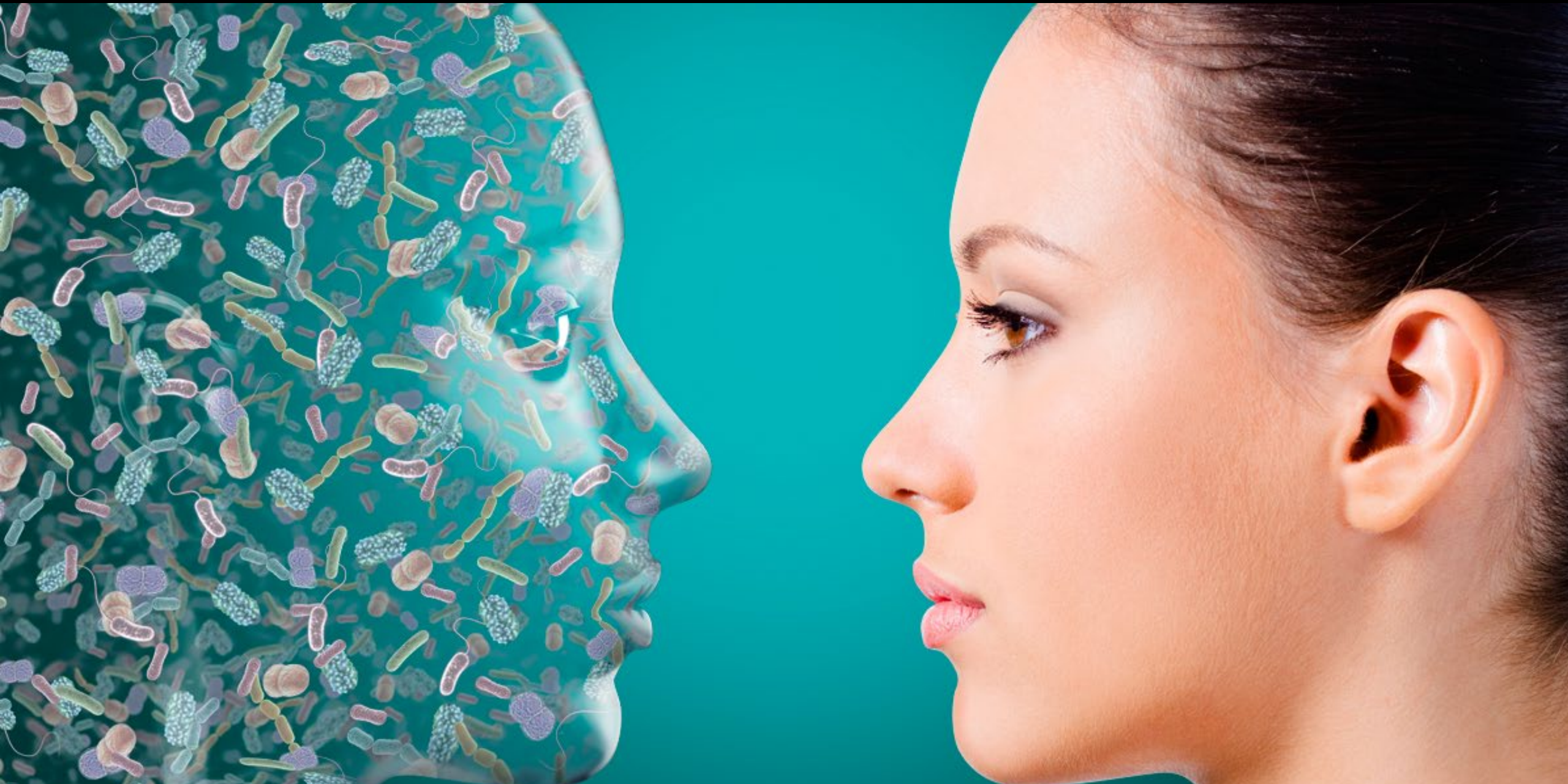


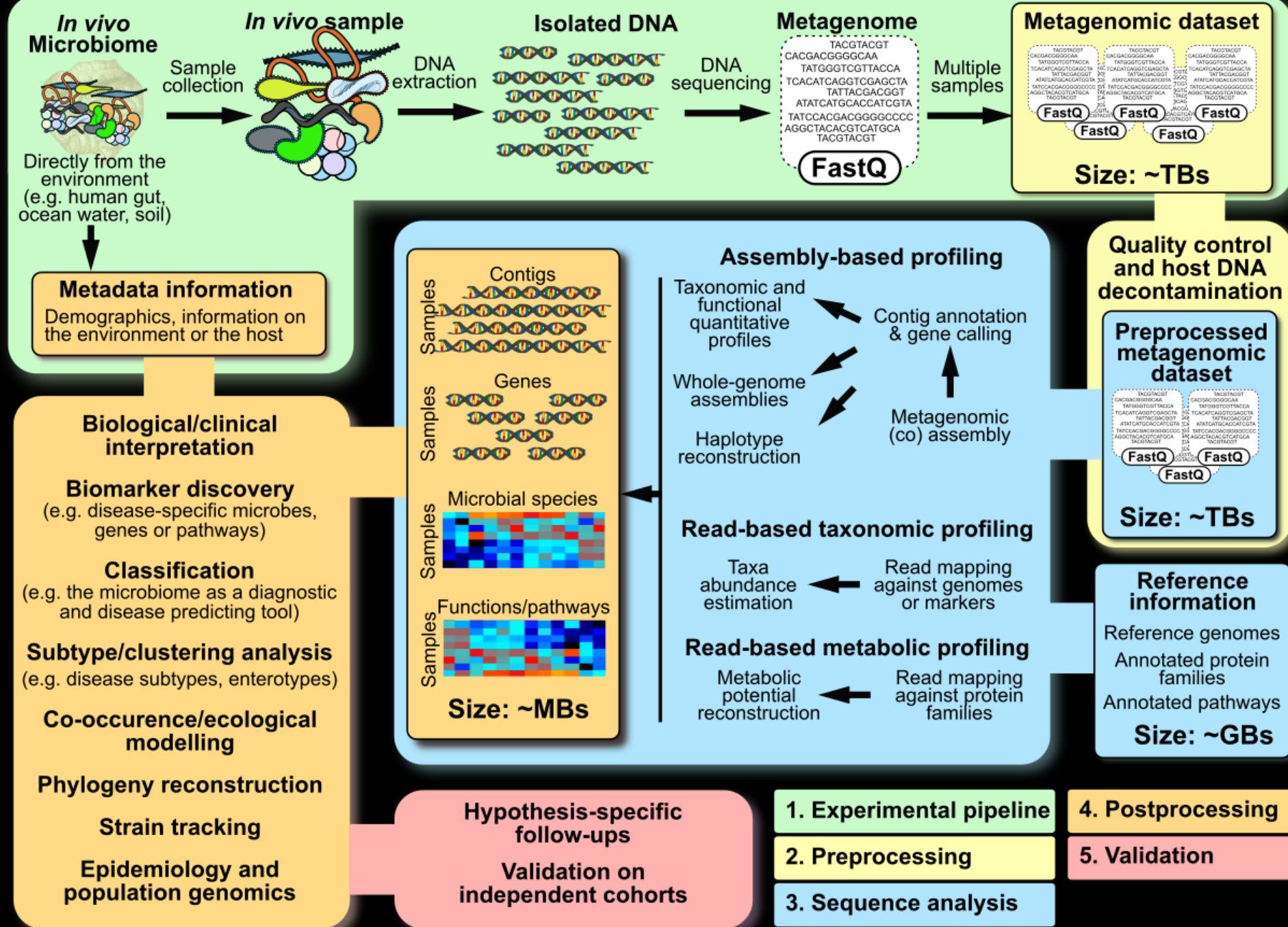
IEO  
Istituto Europeo  
di Oncologia

Department of Cellular, Computational, and  
Integrative Biology (CIBIO). Trento, Italy  
European Institute of Oncology, Milan, Italy



# The human microbiome





# The workflow of shotgun metagenomics

5-10Gb/sample

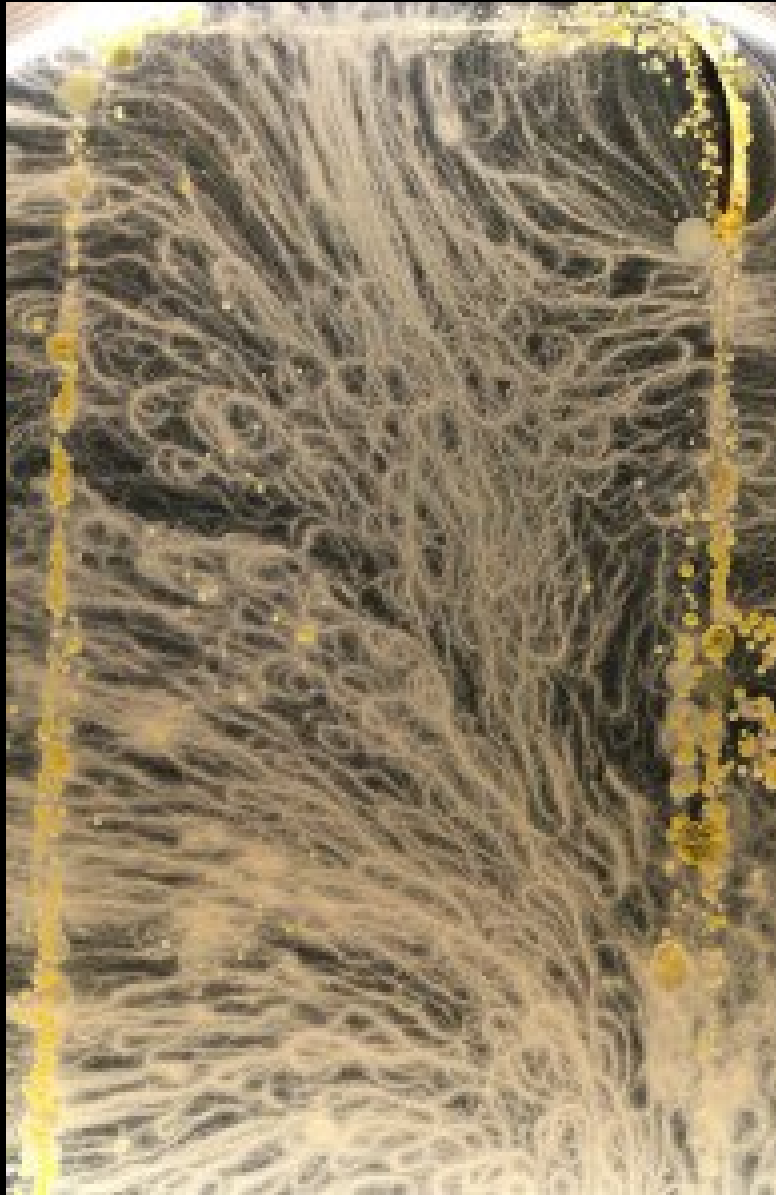
- Our default setting:
- 384 samples on the NovaSeq S4
  - 7.5 Gb/sample

- Human DNA:
- <3% in stool
  - 70%-90% in saliva
  - >95% in skin

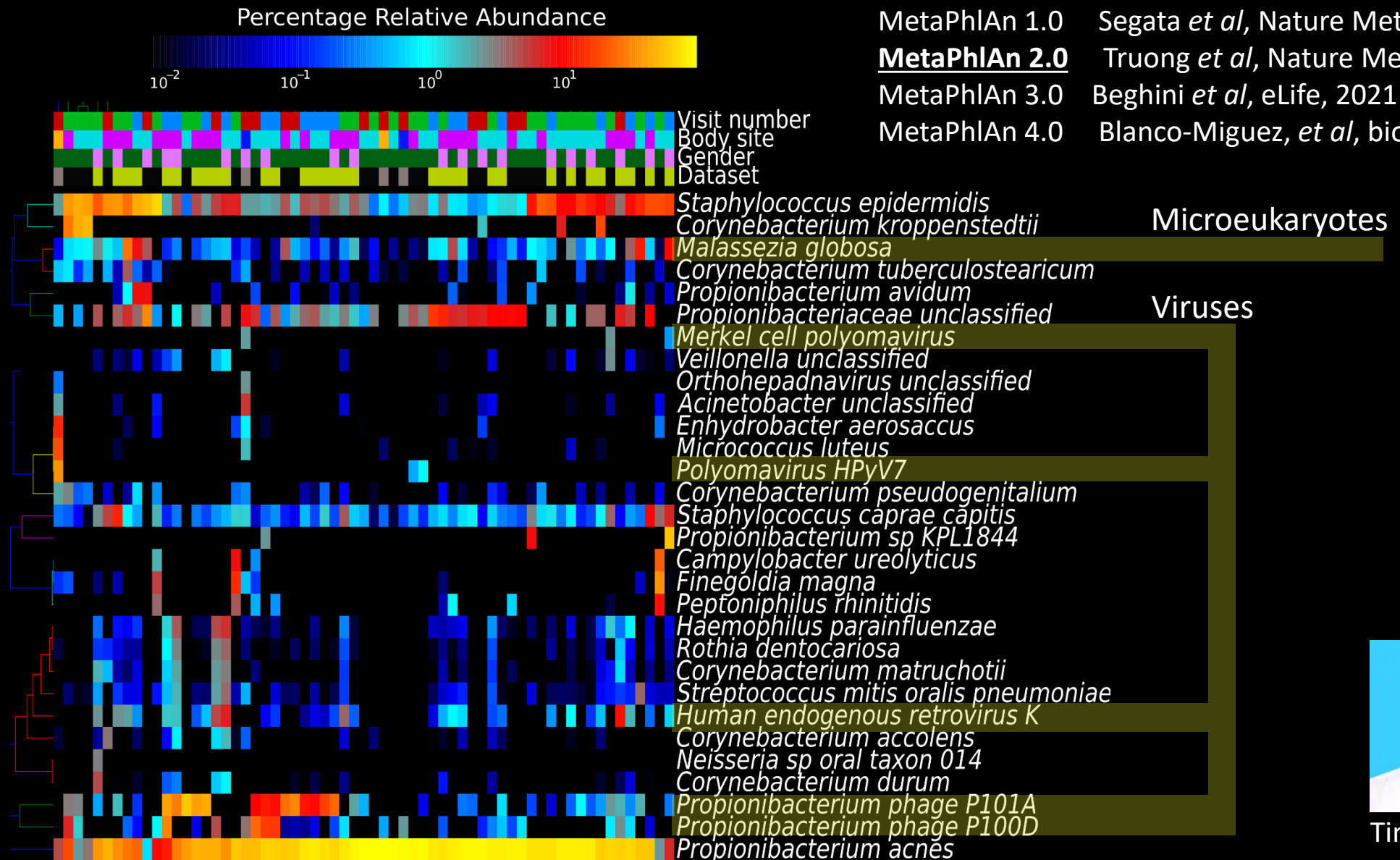
1. Experimental pipeline
2. Preprocessing
3. Sequence analysis
4. Postprocessing
5. Validation

~1% of the phone (or skin?) microbiome

---



# ~60% of the skin microbiome



MetaPhlAn 1.0 Segata *et al*, Nature Methods 2012  
**MetaPhlAn 2.0** Truong *et al*, Nature Methods, 2015  
 MetaPhlAn 3.0 Beghini *et al*, eLife, 2021  
 MetaPhlAn 4.0 Blanco-Miguez, *et al*, bioRxiv 2022






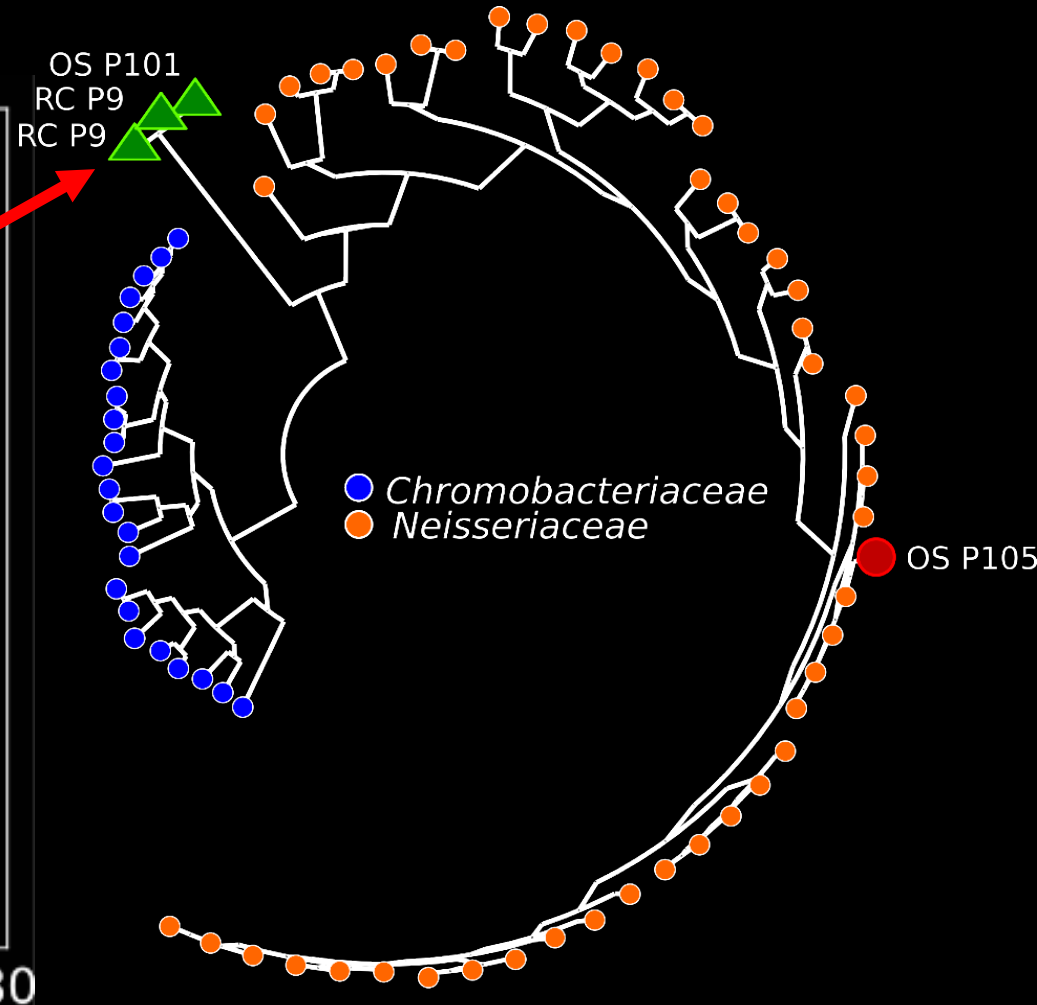
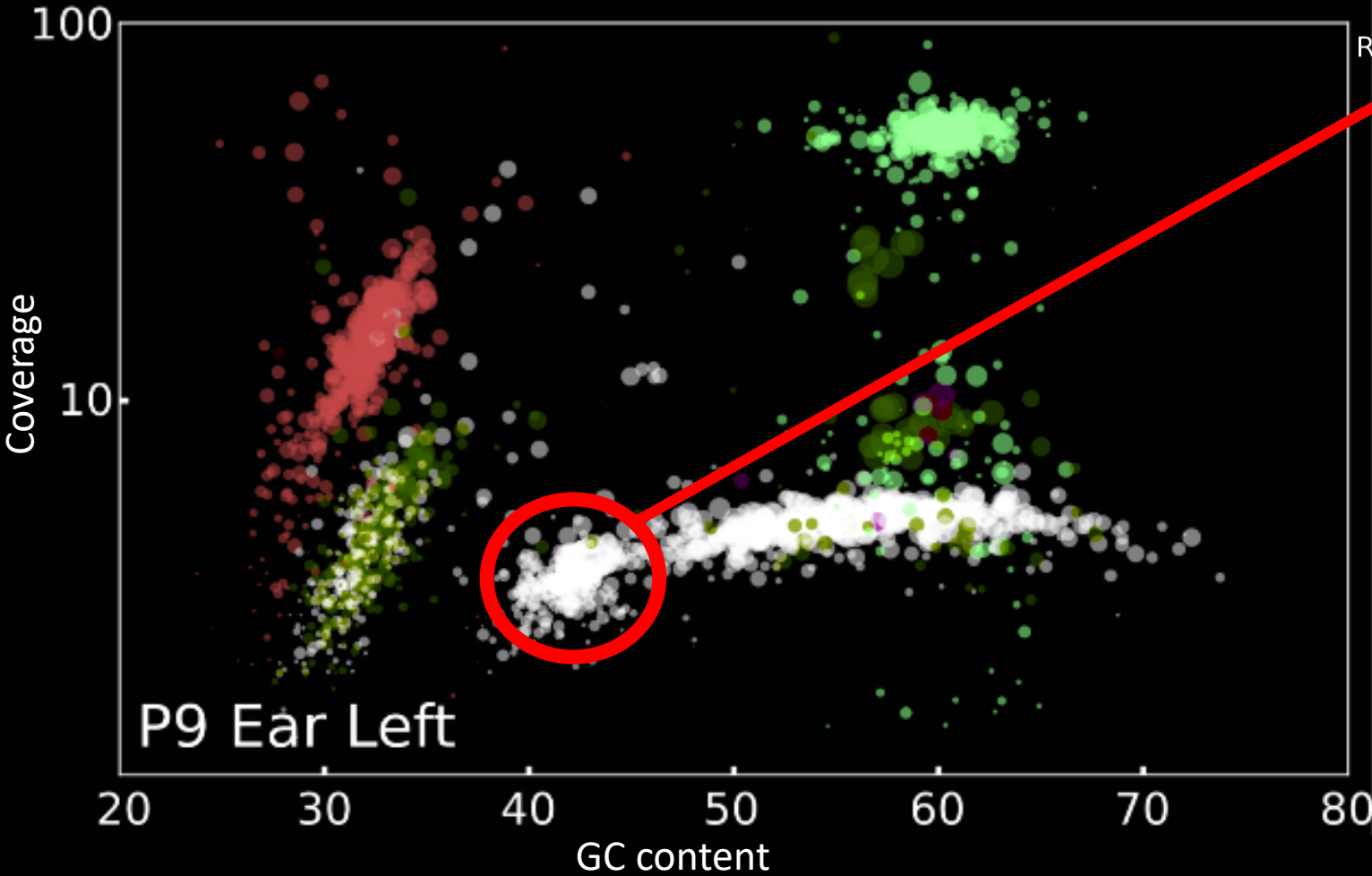
Tin Truong

# The remaining ~40%: the microbial dark matter!

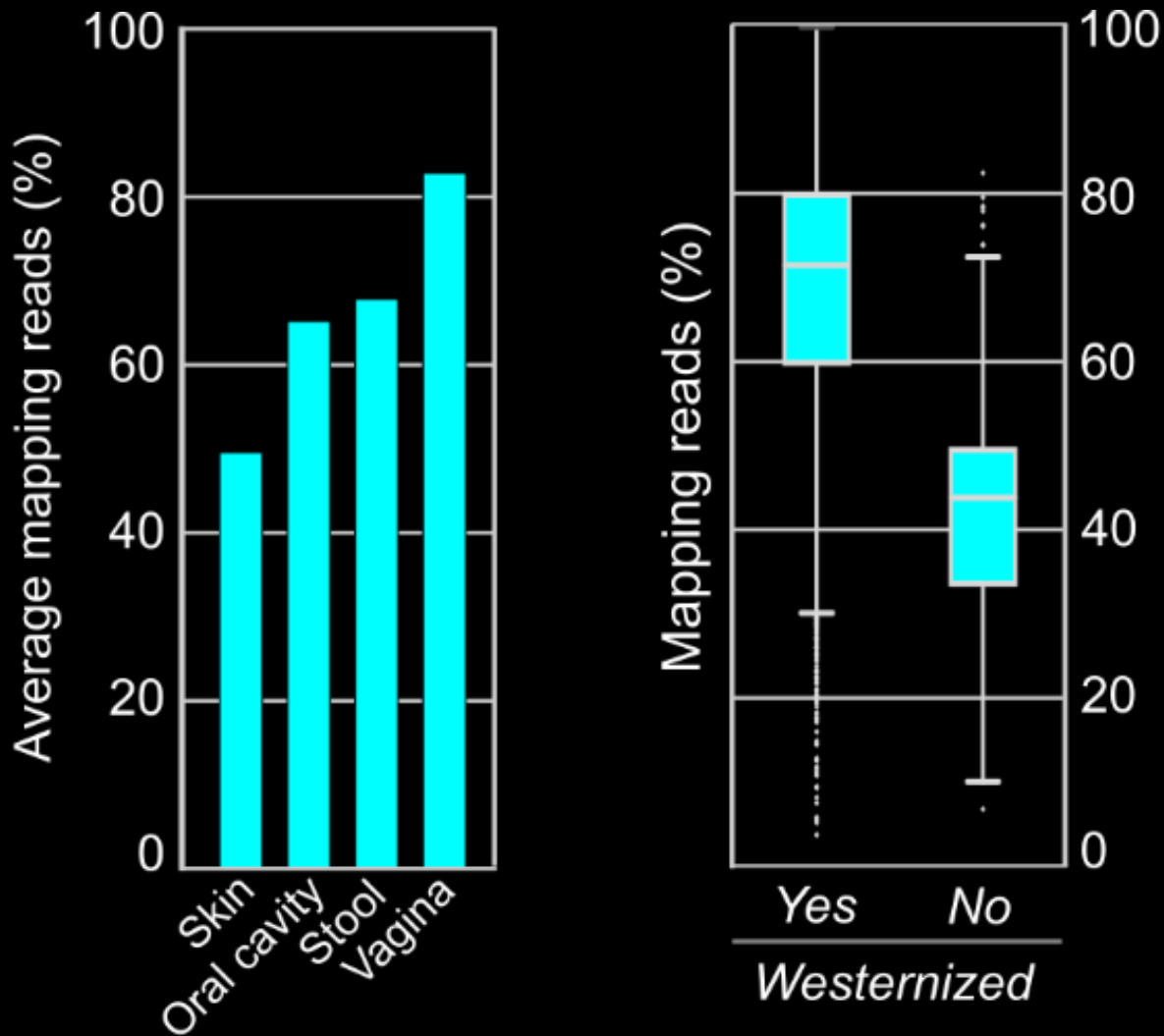
PhyloPhlAn (Segata et al, Nat Comm 2013)

PhyloPhlAn 3 (Asnicar, Nat Comm 2020)

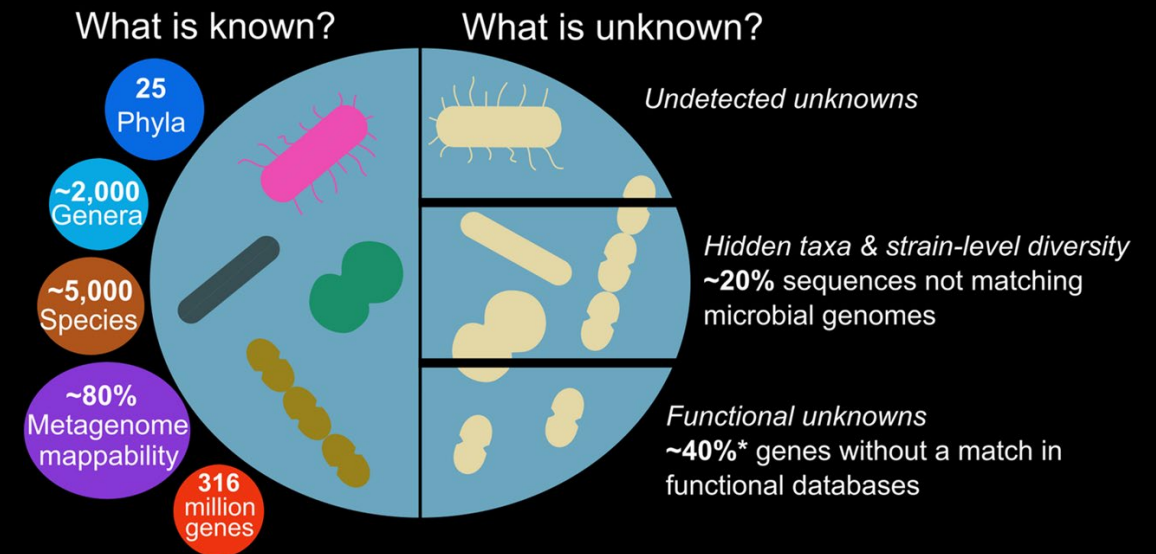
 *Cutibacterium acnes*     *Staphyloccous epidermidis*     Unknowns!



# Many species are still uncharacterized



- Thousands of unknown species/strains?
- Millions of unsampled genes?
- Missing links with diseases/conditions?

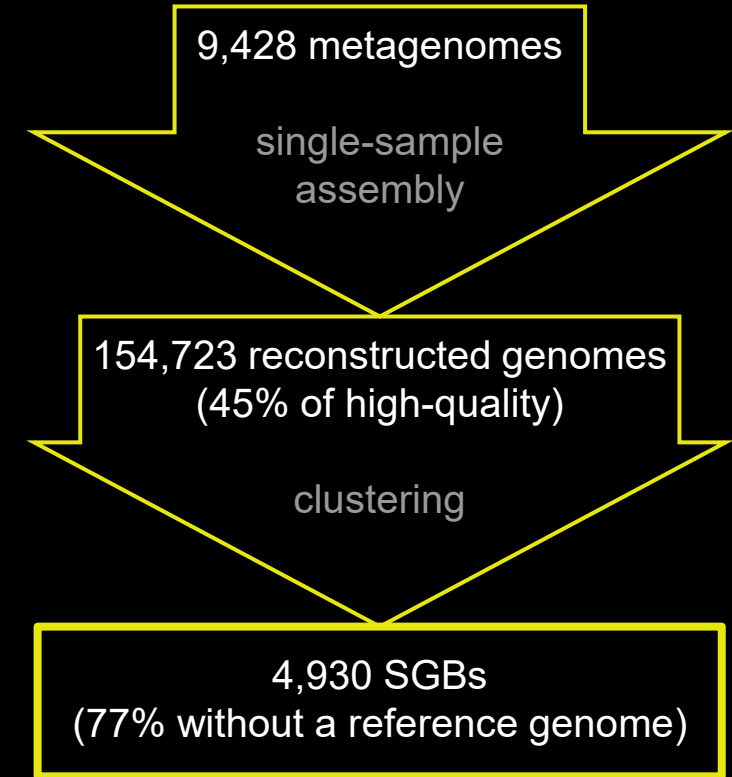
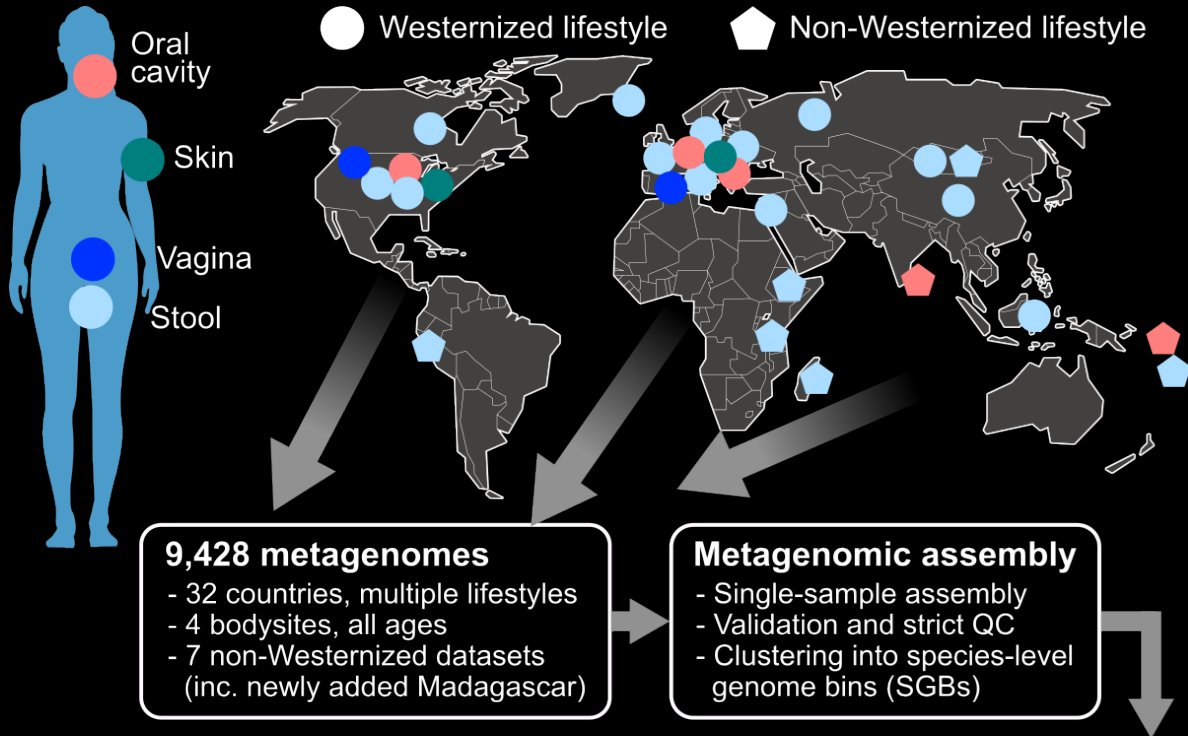


Thomas & Segata, *BMC Biology*, 2019

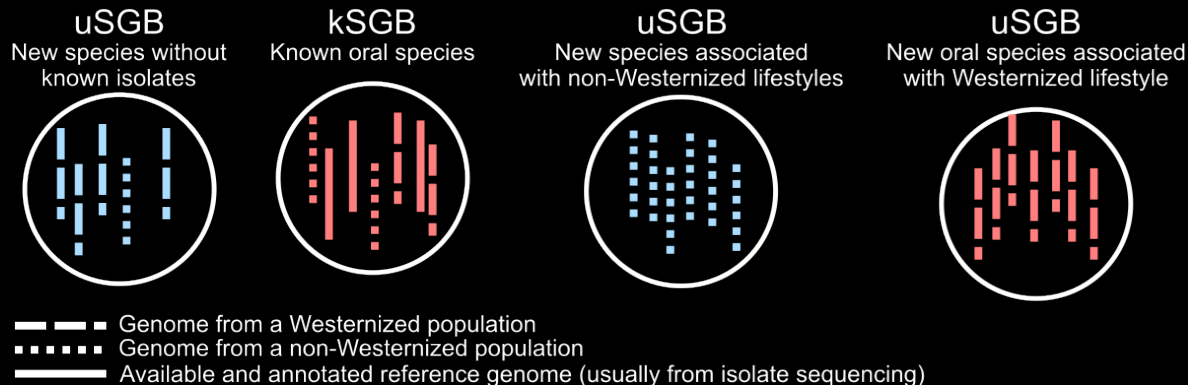


# But there is even more unknown in the human microbiome!

Edoardo Pasolli *et al.*, Cell, 2019



## 154,723 microbial genomes from metagenomes



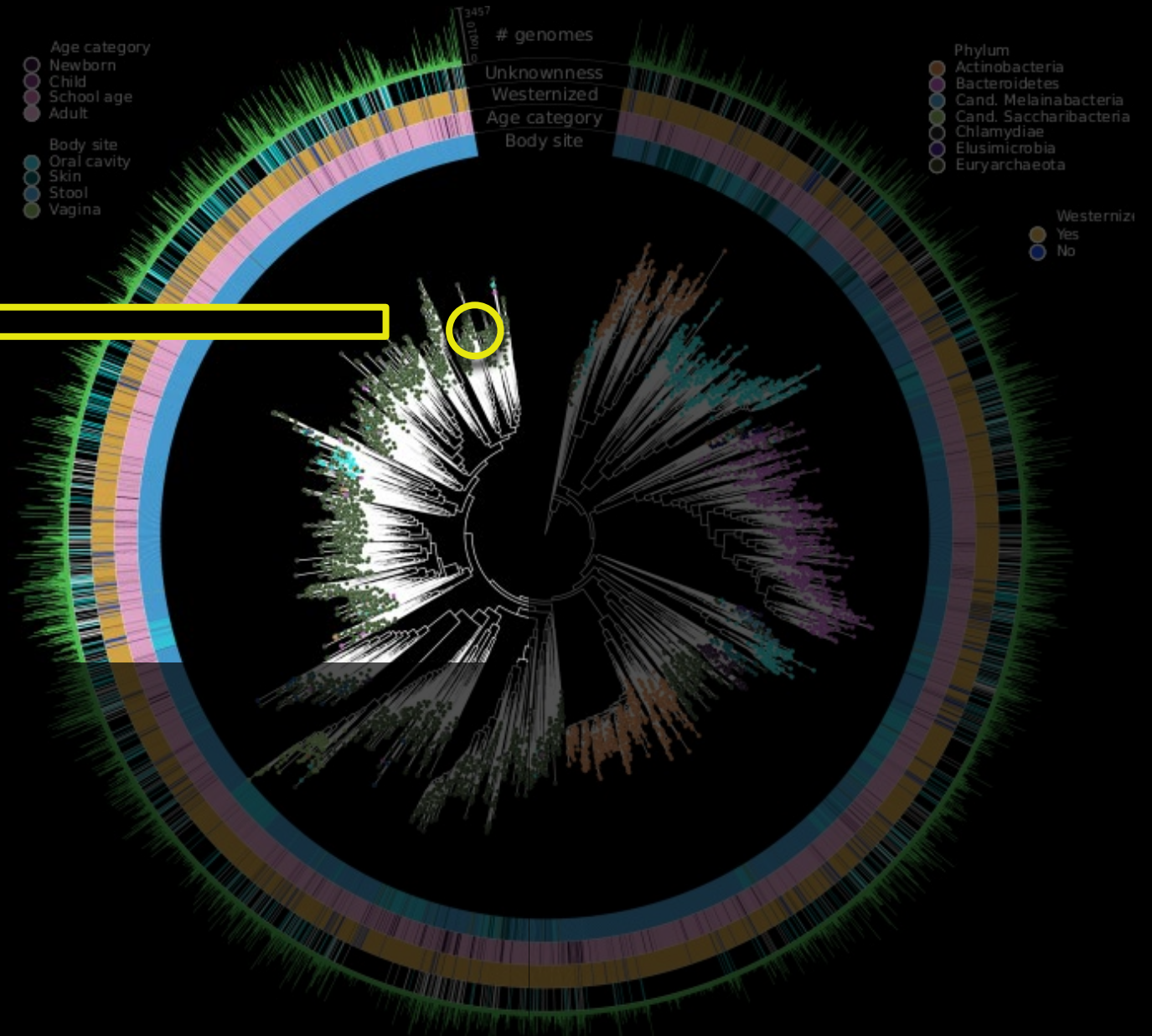
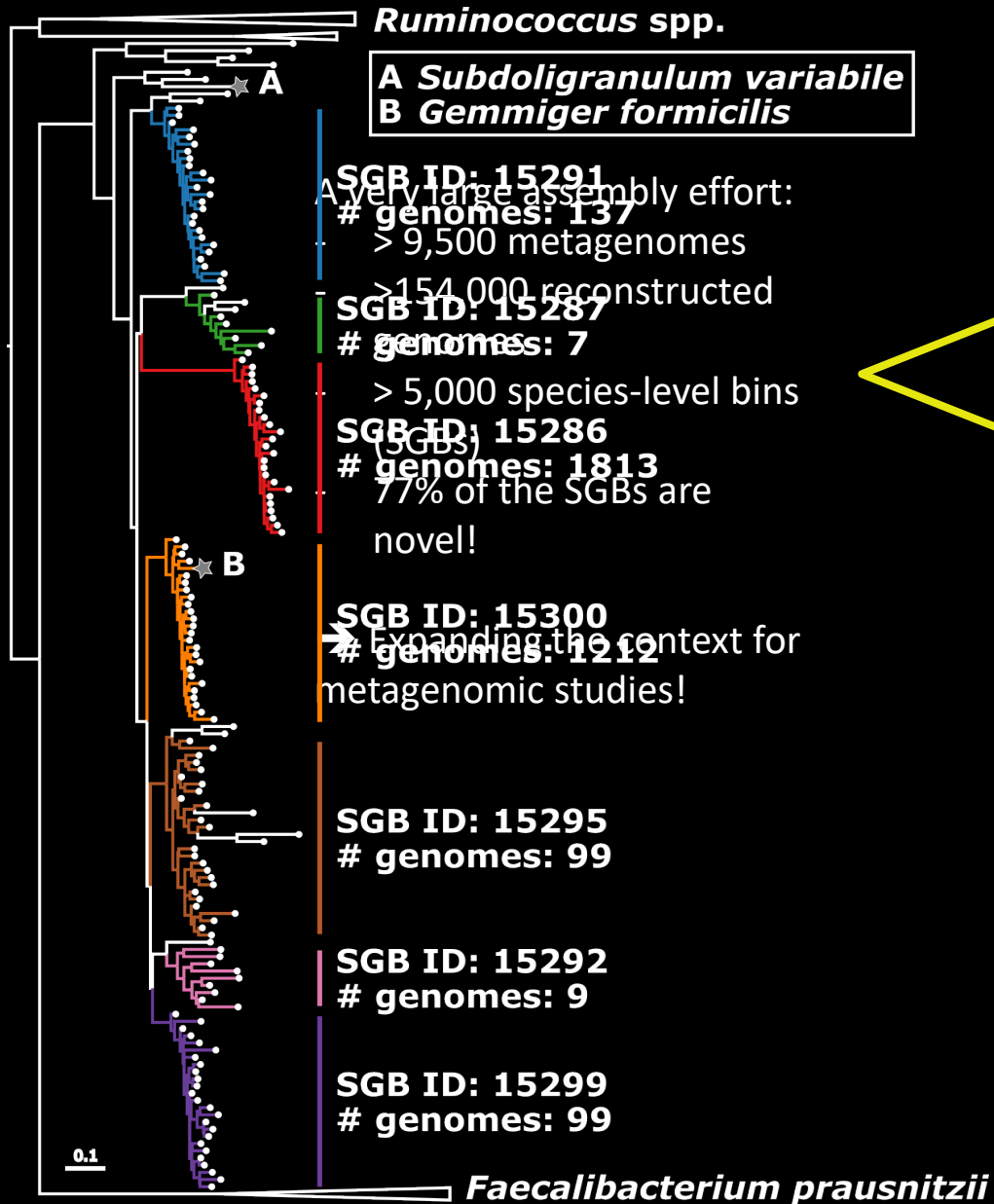
SGB = Species-level Genome Bin

kSGB = known SGB, at least one ref. genome in the cluster

uSGB = unknown SGB, no reference genome in the cluster

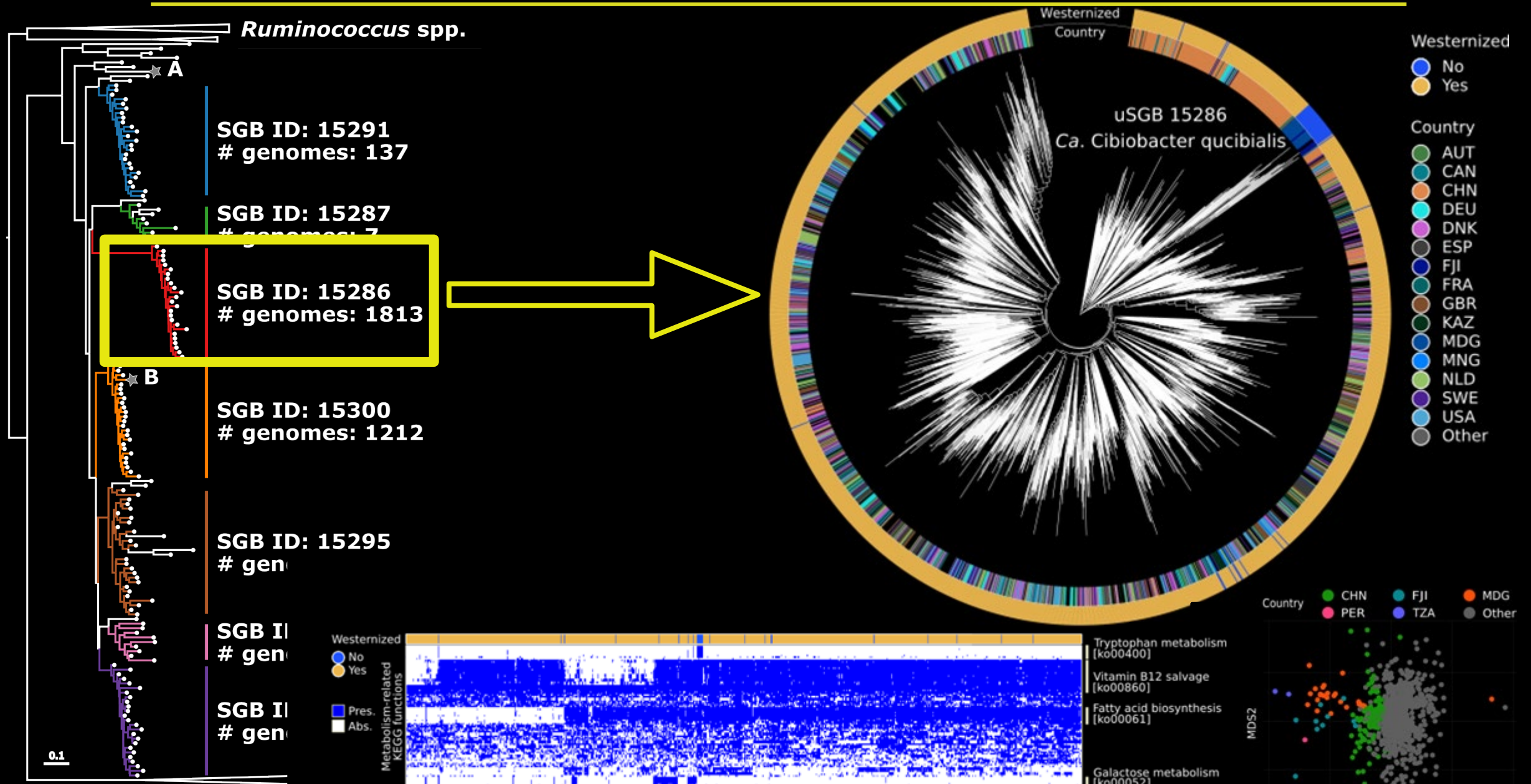
# Reconstructing and cataloguing >150,000 human microbiome genomes

Edoardo Pasoli *et al.*, Cell, 2019



# The seventh most prevalent species: *Cibiobacter qucibialis*

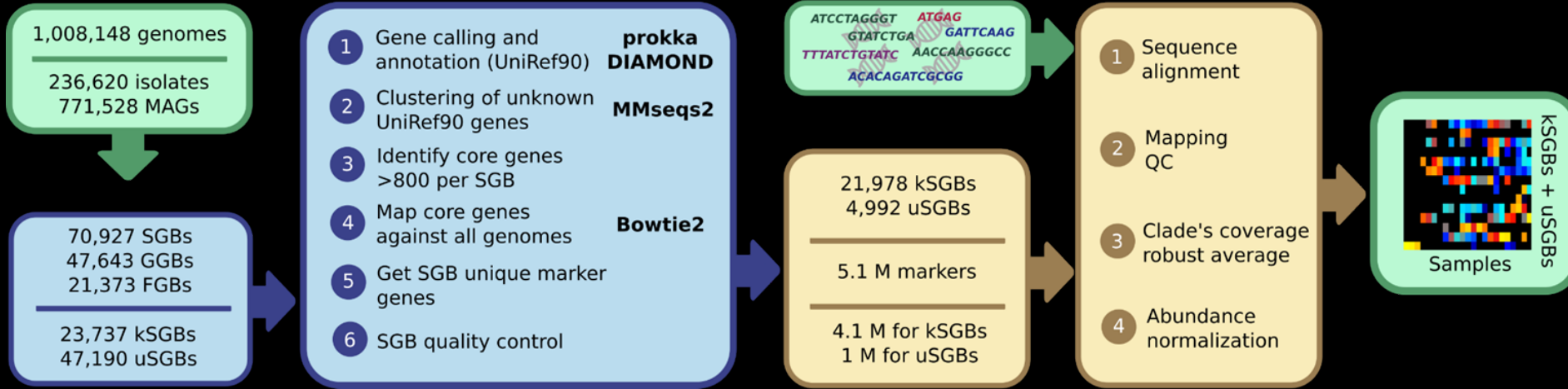
Edoardo Pasoli *et al.*, Cell, 2019



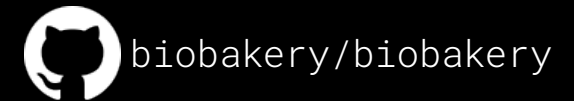
# MetaPhlAn 4: profiling the unknowns



Aitor Blanco-Miguez



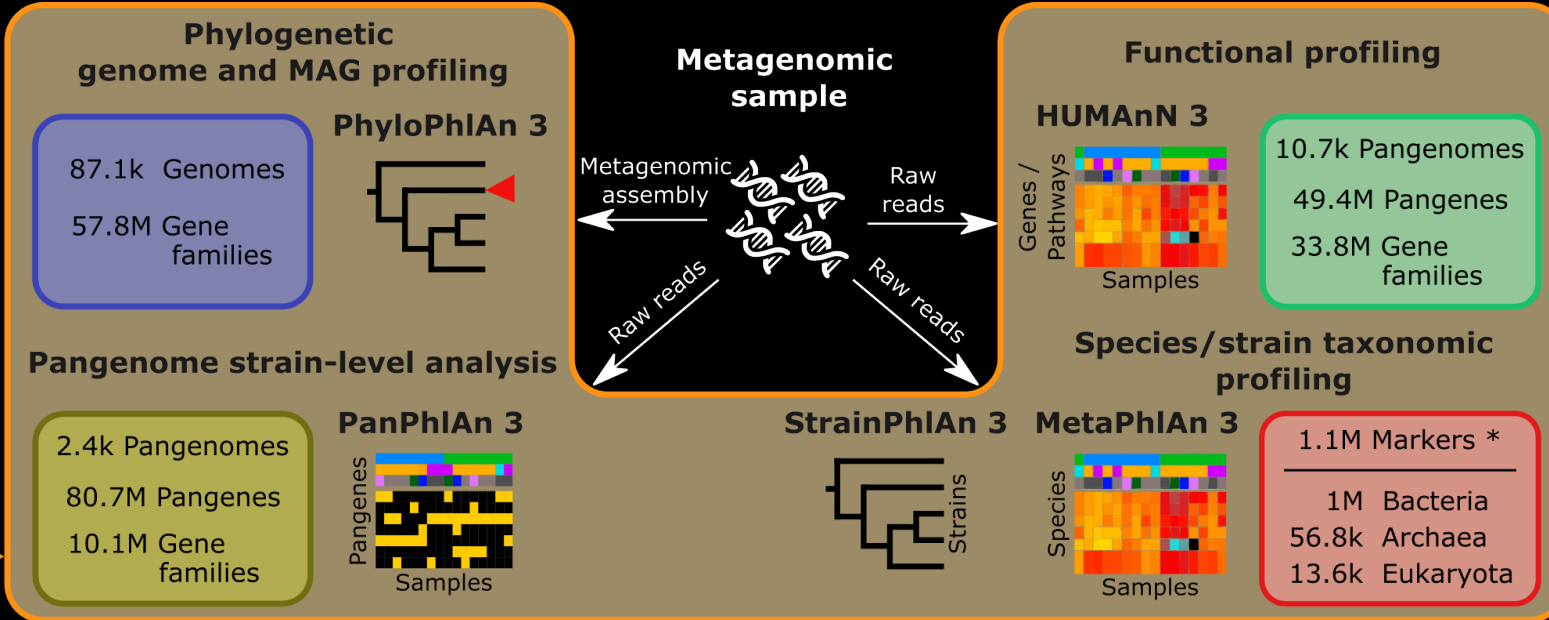
Blanco-Miguez *et al.*, *Nat Biotech*, in revision



## ChocoPhlAn 3

16.8k species  
16k Bacteria  
739 Archaea  
122 Eukaryota

99.2k genomes  
97.9k Bacteria  
947 Archaea  
339 Eukaryota

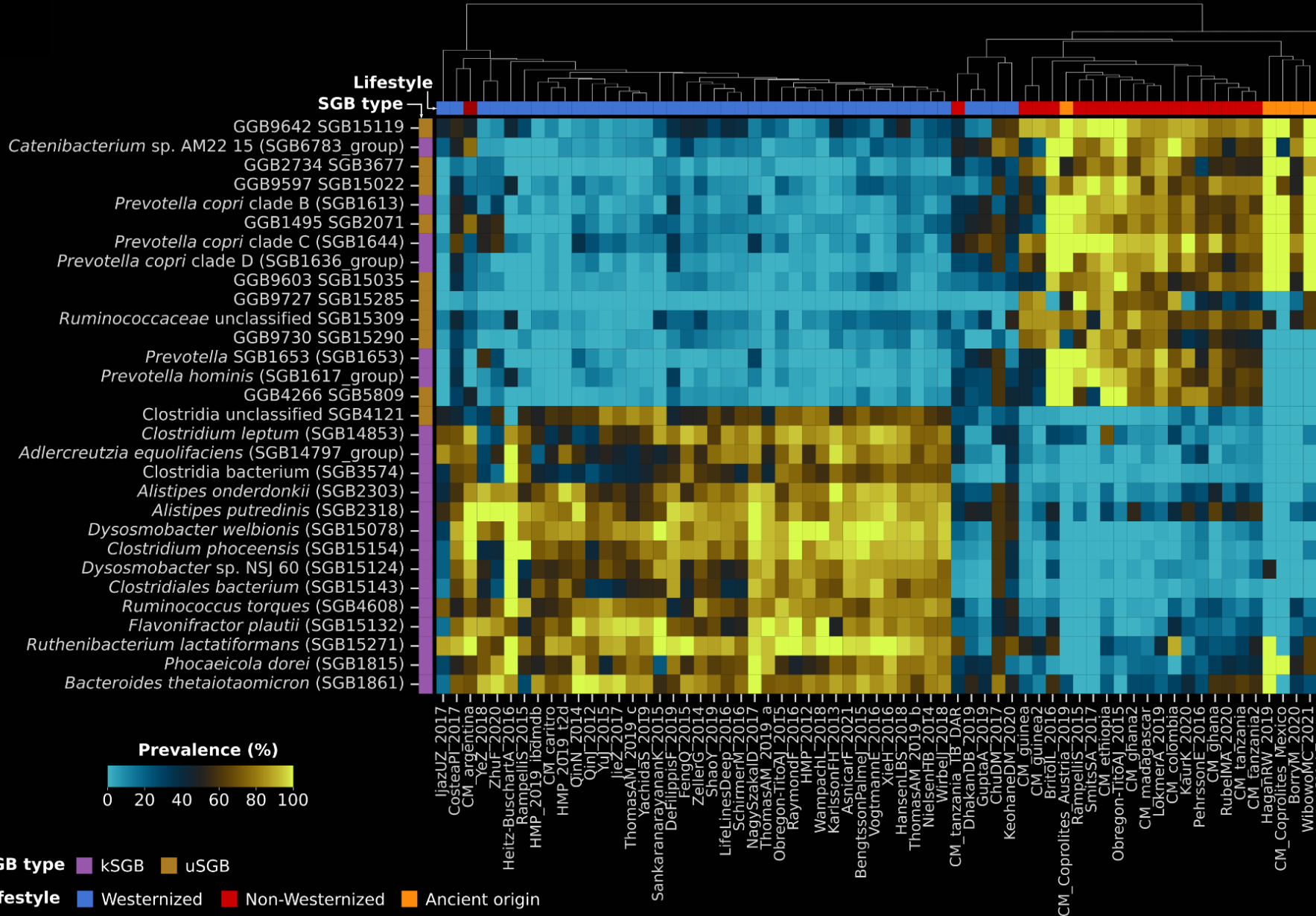


Beghini *et al.*, *eLife*, 2021  
 Karcher *et al.*, *Genome Biology*, 2020  
 Asnicar *et al.*, *Nature Communications*, 2020  
 Zolfo *et al.*, *Nature Biotechnology*, 2019  
 Tett *et al.*, *Cell Host & Microbe*, 2019  
 Pasolli *et al.*, *Cell*, 2019  
 Ferretti *et al.*, *Cell Host&Microbe*, 2018  
 Beghini *et al.*, *ISMEJ*, 2017  
 Truong *et al.*, *Genome Research*, 2017  
 Asnicar *et al.*, *mSystems*, 2017  
 Scholz *et al.*, *Nature Methods*, 2016  
 Ward *et al.*, *Cell Reports*, 2016  
 Donati *et al.*, *Nature Microbiology*, 2016  
 Zolfo *et al.*, *NAR*, 2016  
 Truong *et al.*, *Nature Methods*, 2015.

# Westernized and non-Westernized unknowns



Aitor  
Blanco-Miguez



**Westernized lifestyle**

11,123 metagenomic samples

59 studies

25 countries

3 continents

**Non-Westernized lifestyle**

1,718 metagenomic samples

22 studies

15 countries

4 continents

**Ancient origin**

29 metagenomic samples

7 studies

5,300 to 150 years ago

# Westernized and non-Westernized unknowns



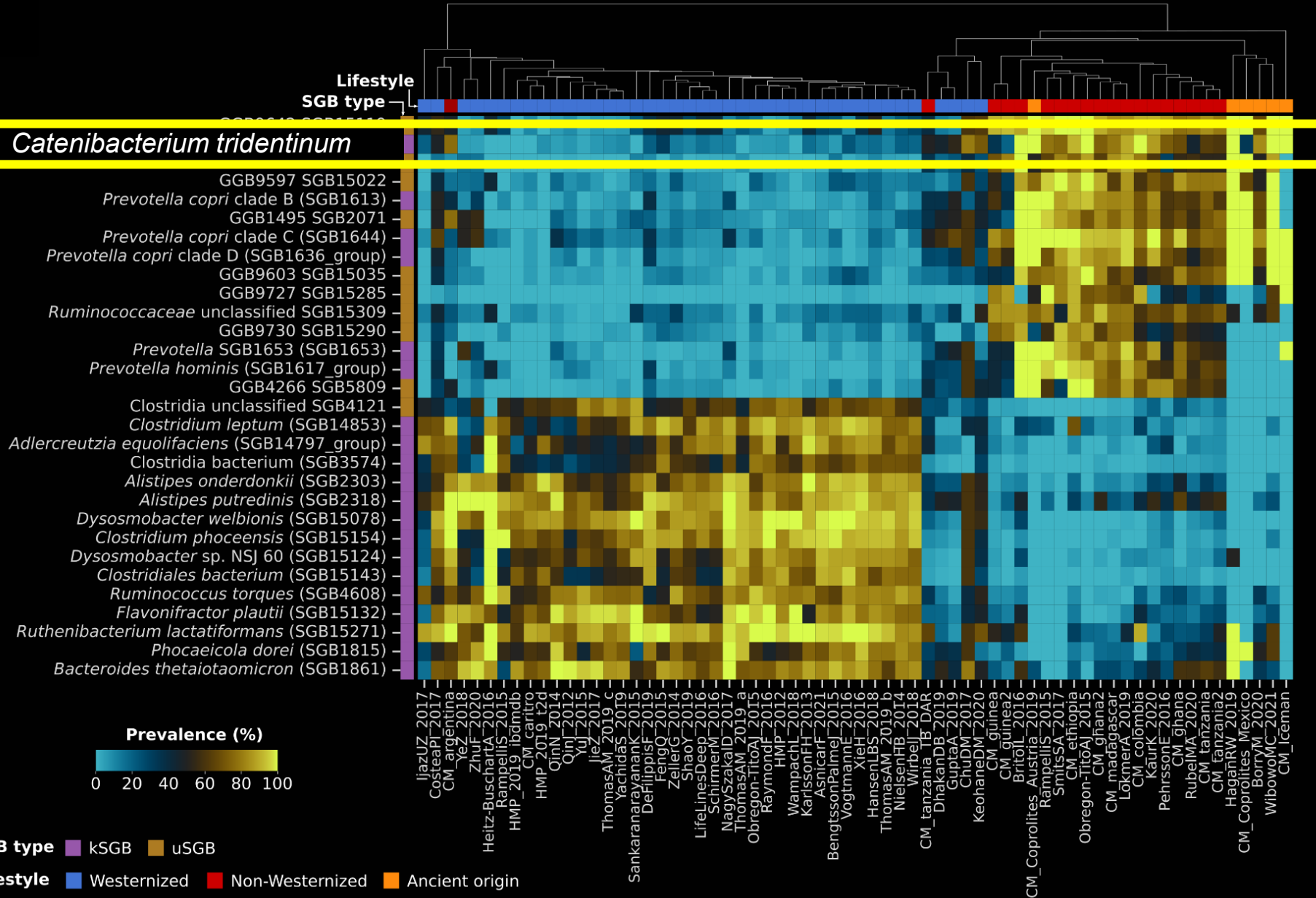
Aitor  
Blanco-Miguez



Liviana  
Ricci



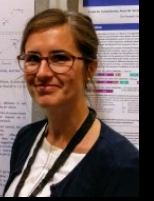
Marta  
Selma-Royo



# Westernized and non-Westernized unknowns



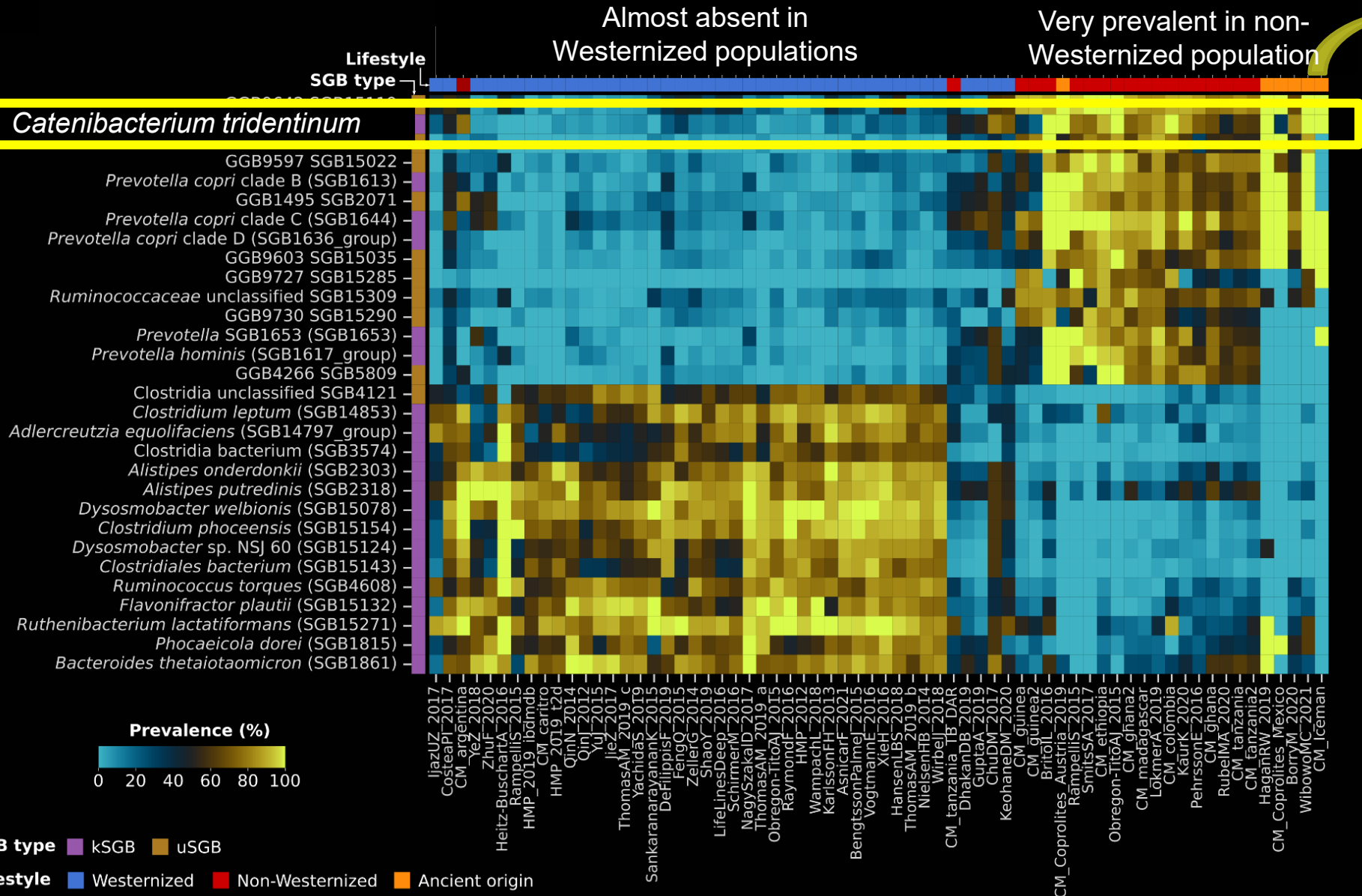
Aitor  
Blanco-Miguez



Liviana  
Ricci



Marta  
Selma-Royo



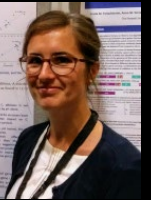
"The Ötzi Iceman"

Found in ancient stool samples

# Westernized and non-Westernized unknowns



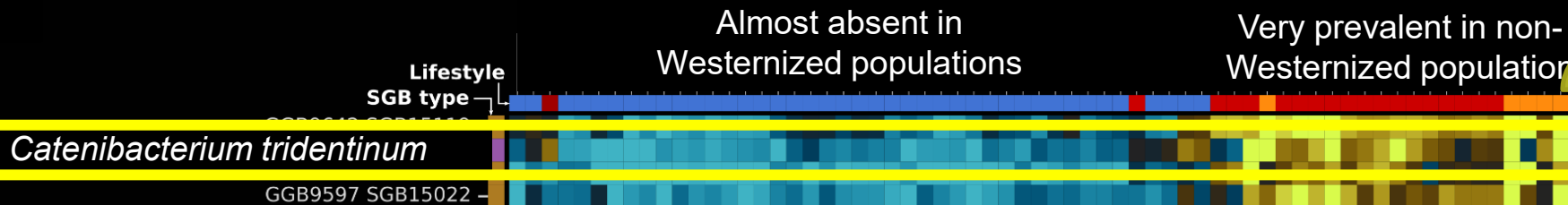
Aitor  
Blanco-Miguez



Liviana  
Ricci



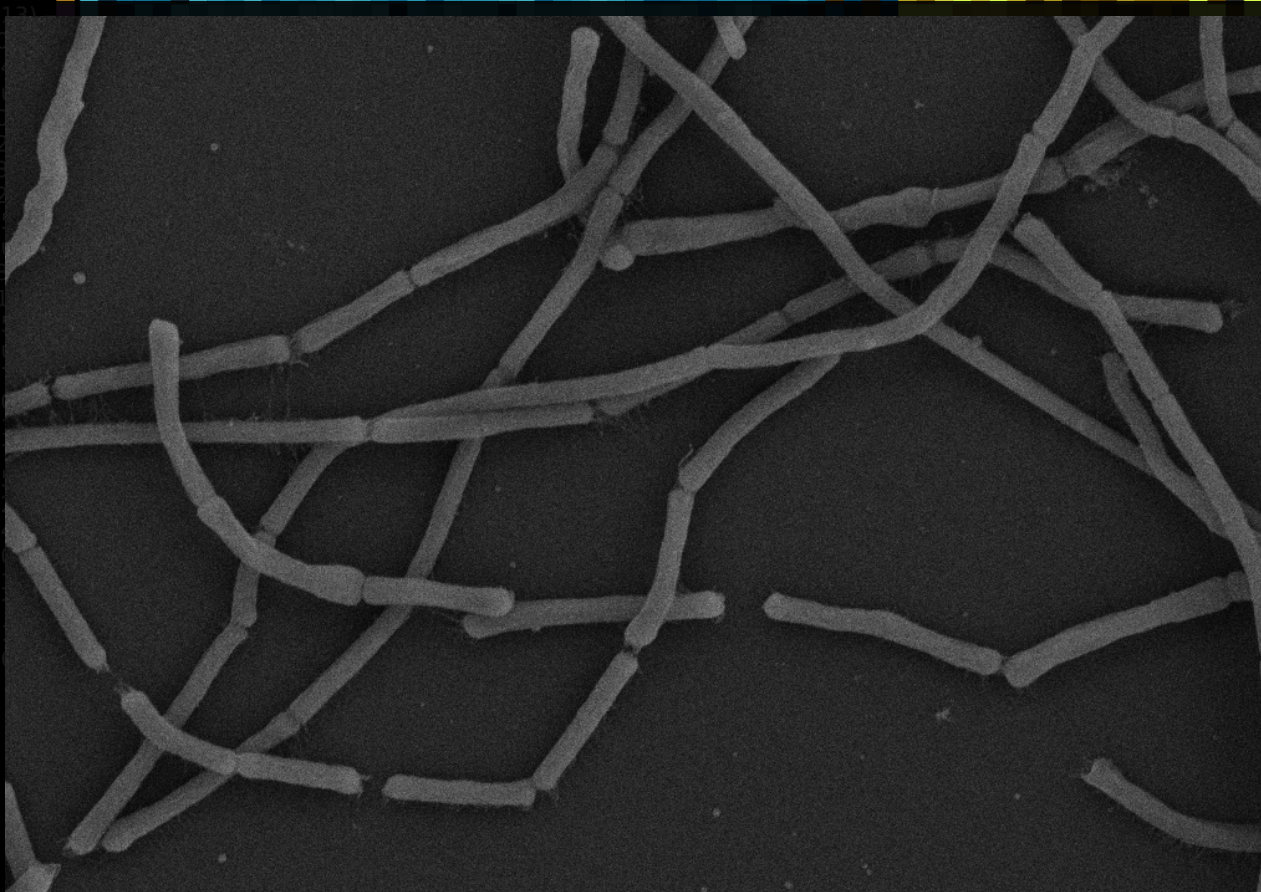
Marta  
Selma-Royo



*Catenibacterium tridentinum*

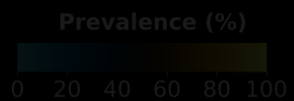
GGB9597 SGB15022

- Prevotella copri clade B (SGB1613)
- GGB1495 SGB20
- Prevotella copri clade C (SGB1616)
- Prevotella copri clade D (SGB1636)
- GGB9603 SGB150
- GGB9727 SGB152
- Ruminococcaceae unclassified (SGB153)
- GGB9730 SGB152
- Prevotella (SGB1653)
- Prevotella hominis (SGB1617)
- GGB4266 SGB58
- Clostridia unclassified (SGB47)
- Clostridium leptum (SGB148)
- Adlercreutzia equolifaciens (SGB14797)
- Clostridia bacterium (SGB35)
- Alistipes onderdonkii (SGB23)
- Alistipes putredinis (SGB23)
- Dysosmobacter welbionis (SGB150)
- Clostridium phoceensis (SGB151)
- Dysosmobacter sp. NSJ 60 (SGB151)
- Clostridiales bacterium (SGB151)
- Ruminococcus torques (SGB46)
- Flavonifractor plautii (SGB151)
- Ruthenibacterium lactatiformans (SGB152)
- Phocaeicola dorei (SGB18)
- Bacteroides thetaiotaomicron (SGB18)



Found in ancient stool samples

\* ... and found + isolated from an anonymous PI in the lab of Computational Metagenomics



SGB type ■ kSGB ■ uSGB  
Lifestyle ■ Westernized ■ Non-Westernized

1 μm

EHT = 6.00 kV  
WD = 7.9 mm

Signal A = SE2  
Mag = 10.00 K X





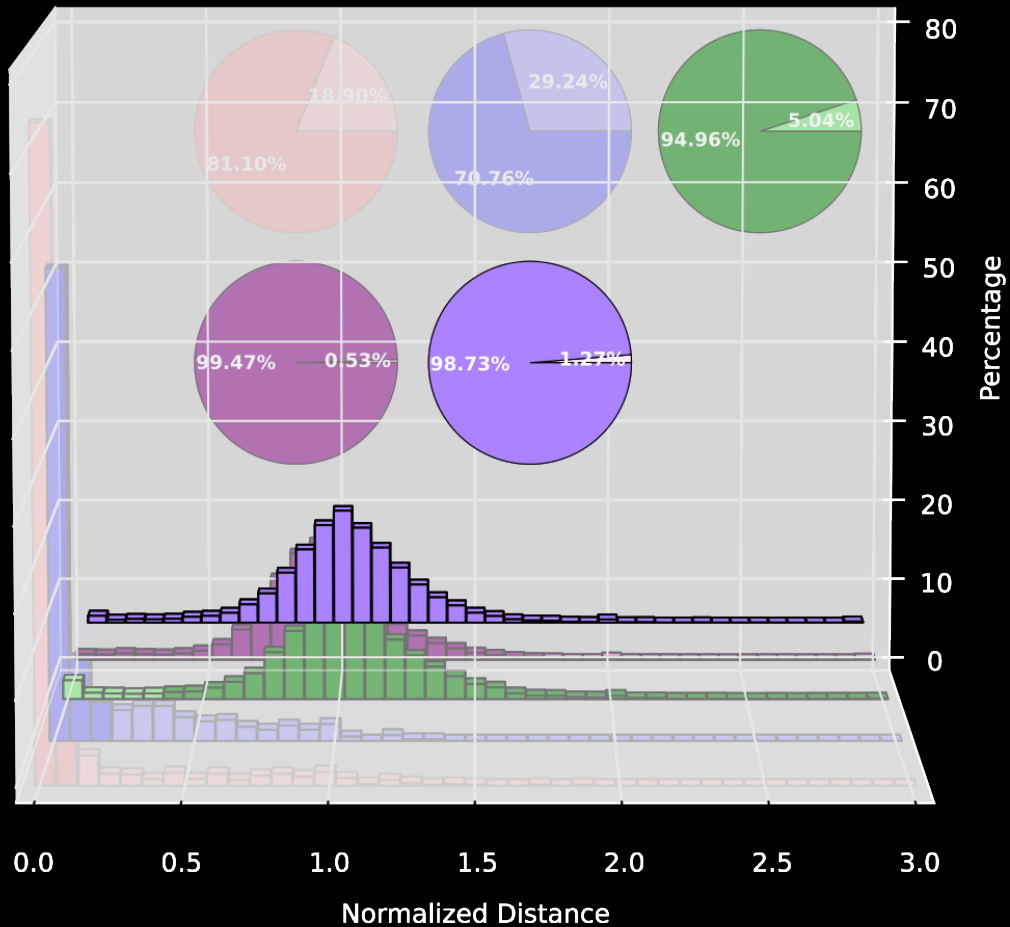
# Each human microbiome is unique at the strain level

- Subjects from around the world (~3000 sbj from 4 continents)
- Subjects from EU (6 countries)
- Subjects from US (from two universities)

Samples from same subjects collected at ~6 months

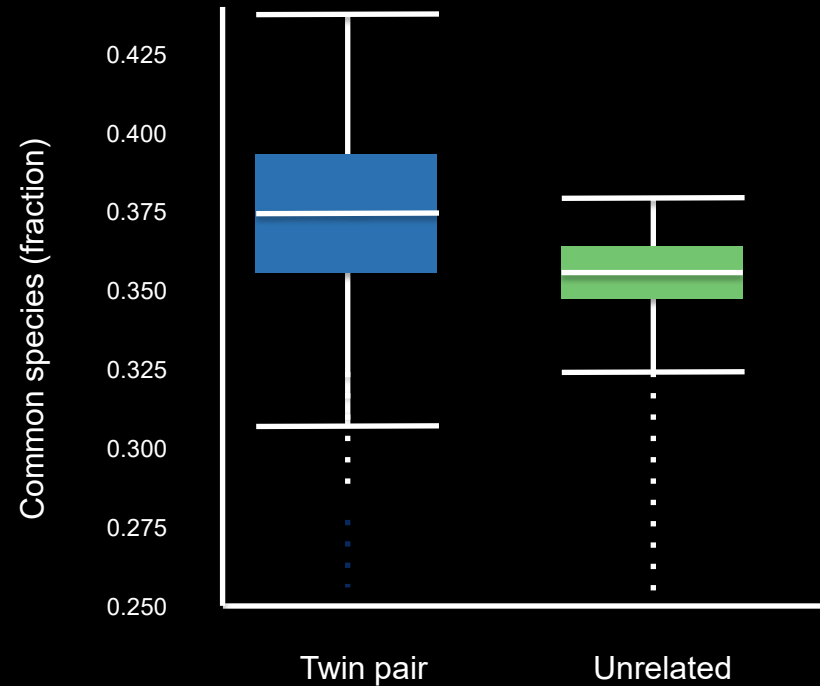
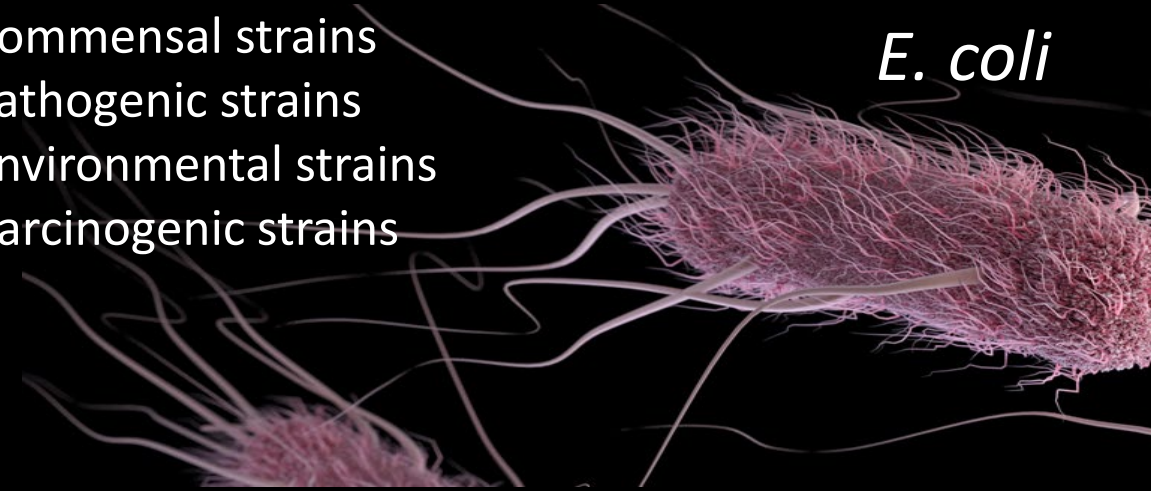
- Subjects from EU
- Subjects from US

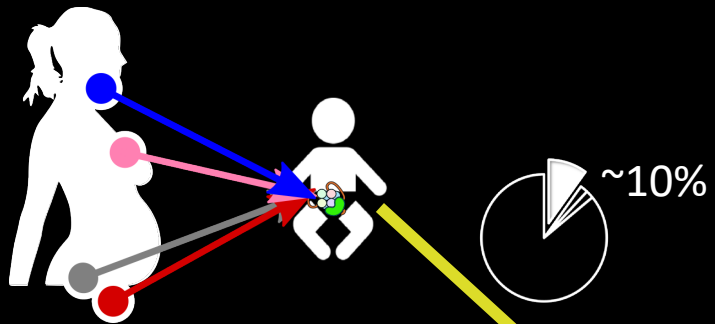
Truong *et al.*, *Genome Research*, 2017



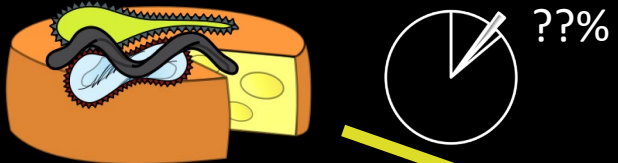
Commensal strains  
Pathogenic strains  
Environmental strains  
Carcinogenic strains

*E. coli*

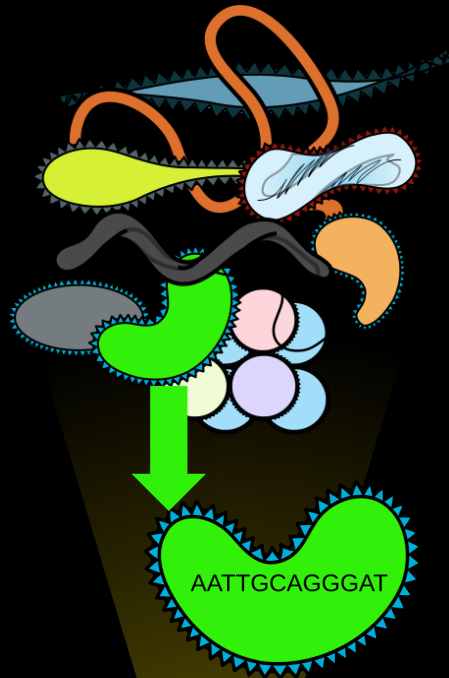




[Ferretti et al, Cell Host & Microbe, 2018  
Valles-Colomer et al, Nature, 2023]

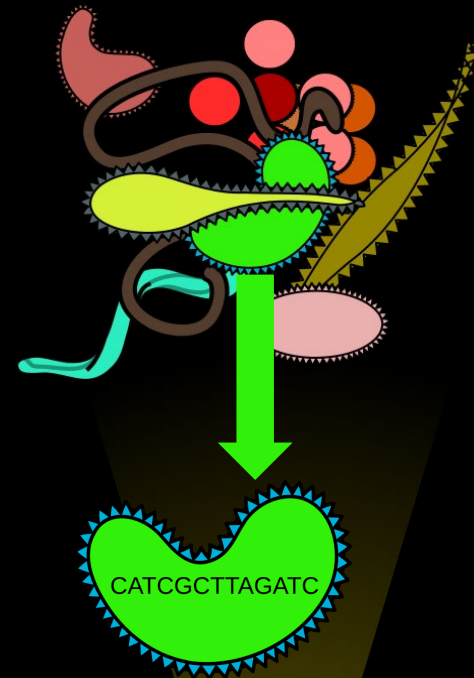


[Pasolli et al, Nature Communications, 2020  
Carlino et al, submitted]



Average 35% shared species  
[Asnicar et al, Nature Medicine, 2021]

< 0.1% of shared strains  
[Truong et al, Genome Research, 2017]  
[Beghini et al, eLife, 2021]  
[Pasolli et al, Cell, 2019]



# Dietary microbial species in the human microbiome

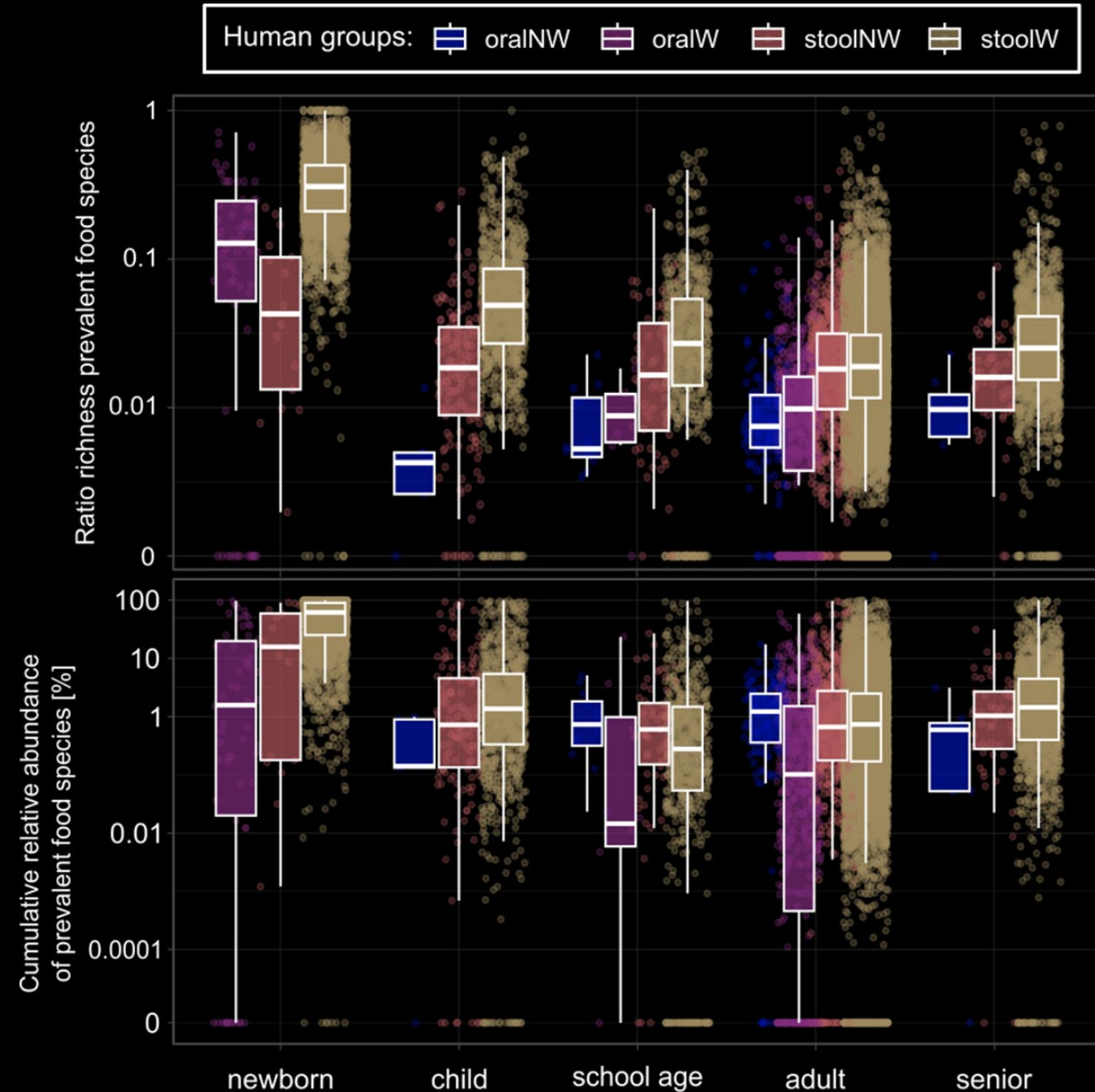
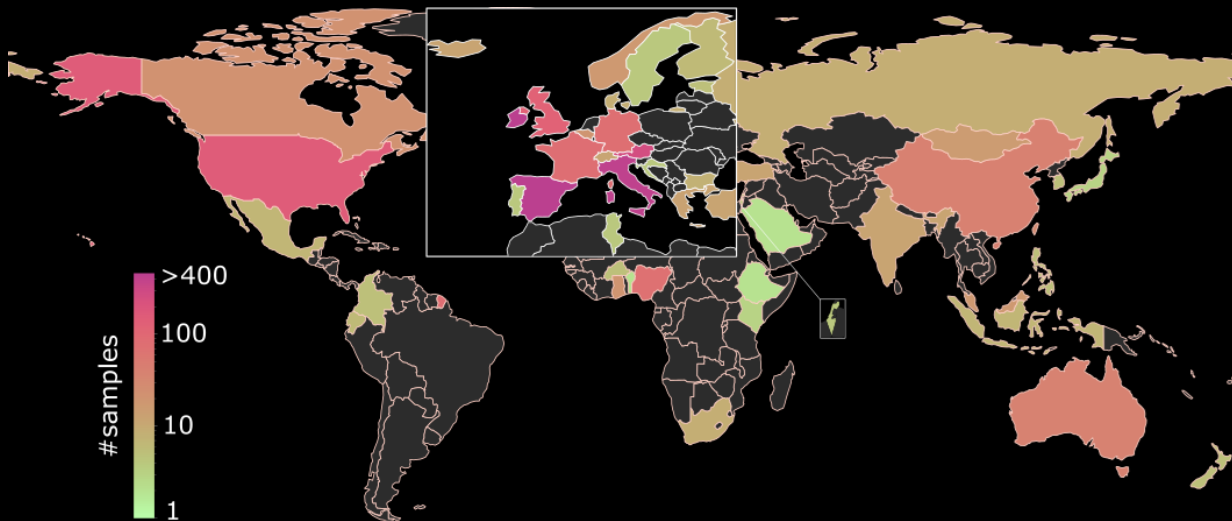


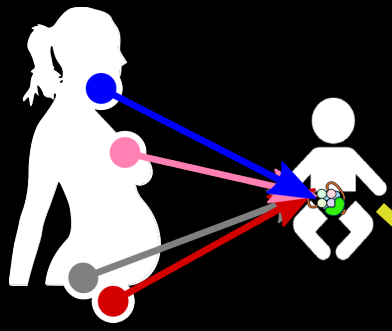
Niccolò  
Carlino



Overlap of 2.5k food and 20k human microbiomes depends on host features:

In adults the food-associated species constituted about 3% of the total microbial population.

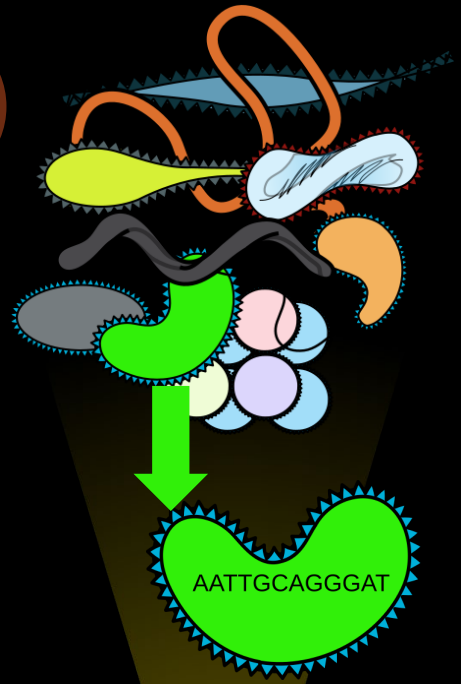




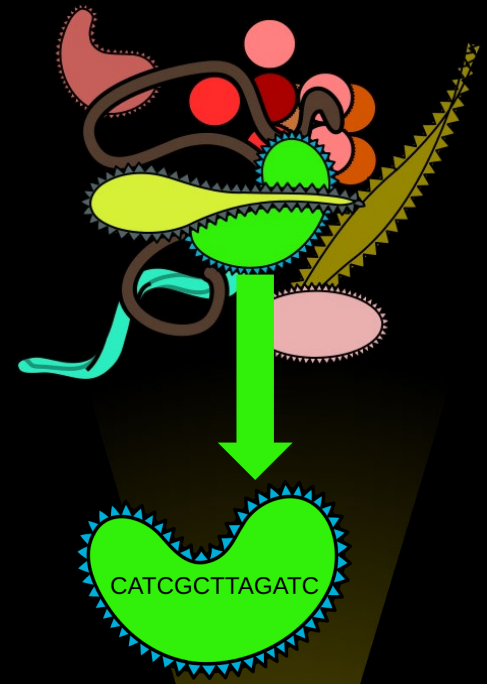
[Ferretti et al, Cell Host & Microbe, 2018  
Valles-Colomer et al, Nature, 2023]



4



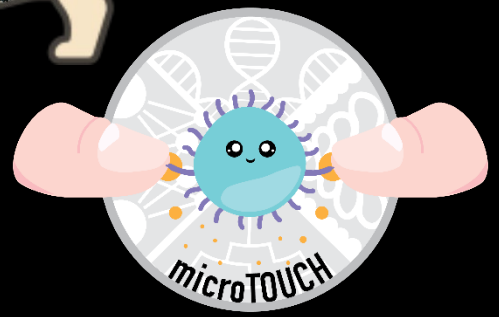
Average 35% shared species  
[Asnicar et al, Nature Medicine, 2021]



< 0.1% of shared strains  
[Truong et al, Genome Research, 2017]  
[Beghini et al, eLife, 2021]  
[Pasolli et al, Cell, 2019]



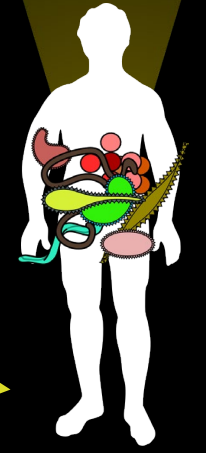
[Pasolli et al, Nature Communications, 2020  
Carlino et al, submitted]



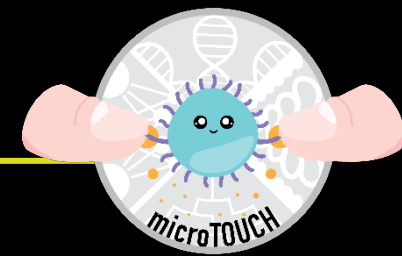
microTOUCH



Hypothesis: extensive and intricate horizontal transmission



# On the transmission of the human microbiome

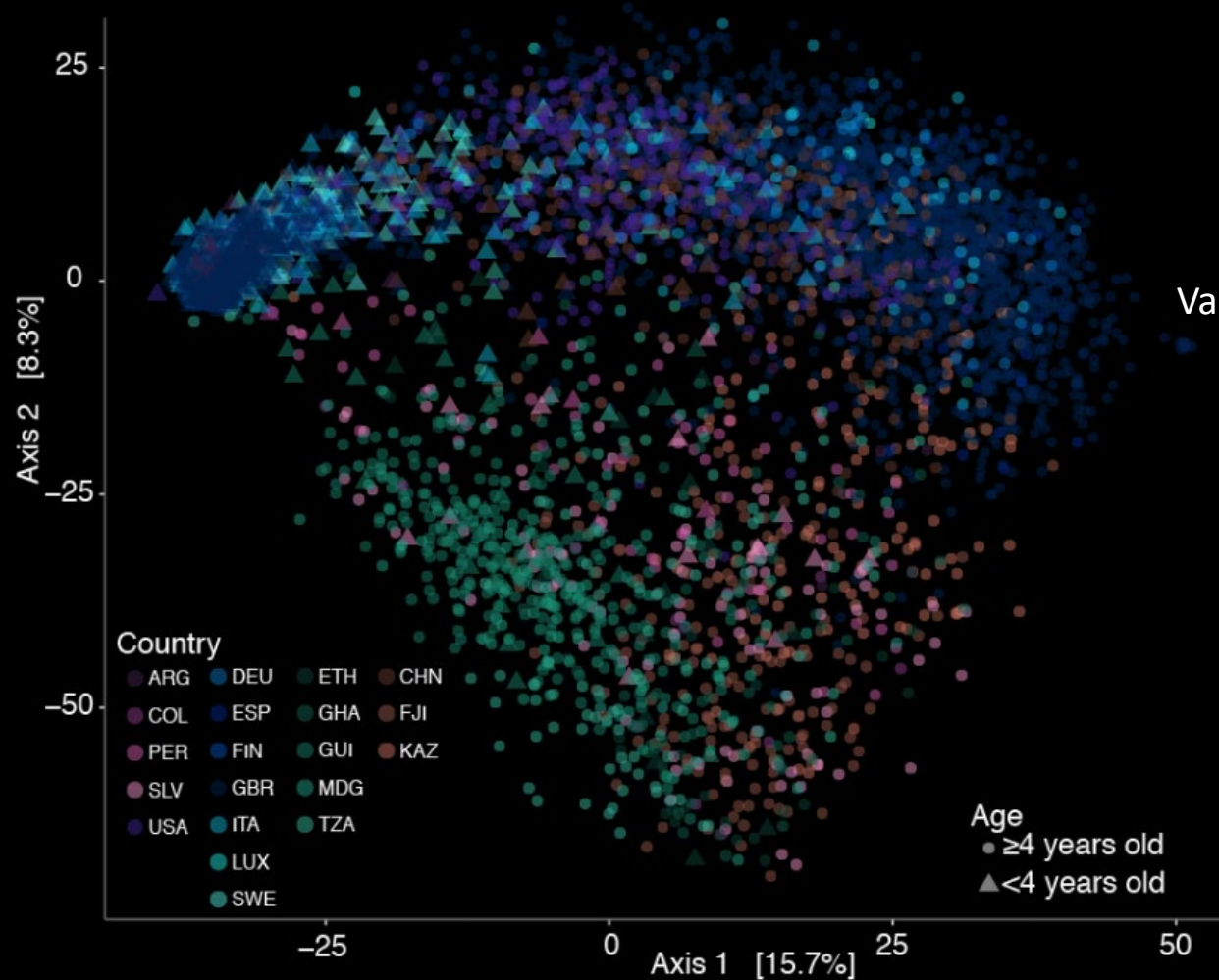


Microbiome transmission set (N=9,715 [2,880])

Gut	Longitudinal sets	Mother-offspring pairs
	1,676 samples [0] 495 individuals [0] 5 datasets [0] 4 countries [0]	3,598 samples [636] 2,144 individuals [593] 711 pairs [261] 18 datasets [8] 14 countries [3]
Oral	Households	Adult twins
	1,123 samples [435] 952 individuals [435] 211 households [70] 7 datasets [3] 8 countries [1]	1,734 samples [0] 1,734 individuals [0] 712 pairs [0] 3 datasets [0] 1 country [0]
Oral	Mother-offspring pairs	Households
	1,315 samples [1,286] 1,315 individuals [1,286] 668 pairs [653] 2 datasets [1] 2 countries [1]	2,069 samples [1,929] 2,069 individuals [1,929] 730 households [646] 2 datasets [1] 2 countries [1]

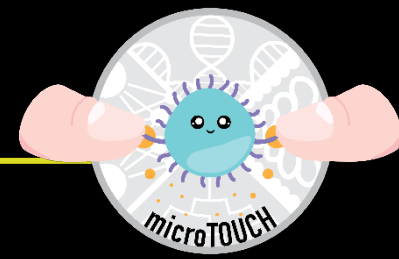
Strain-level metagenomic profiling (StrainPhlAn)

Gut	646 SGBs (365 uSGBs) ➤	477,141 strains
Oral	252 SGBs (60 uSGBs) ➤	326,403 strains



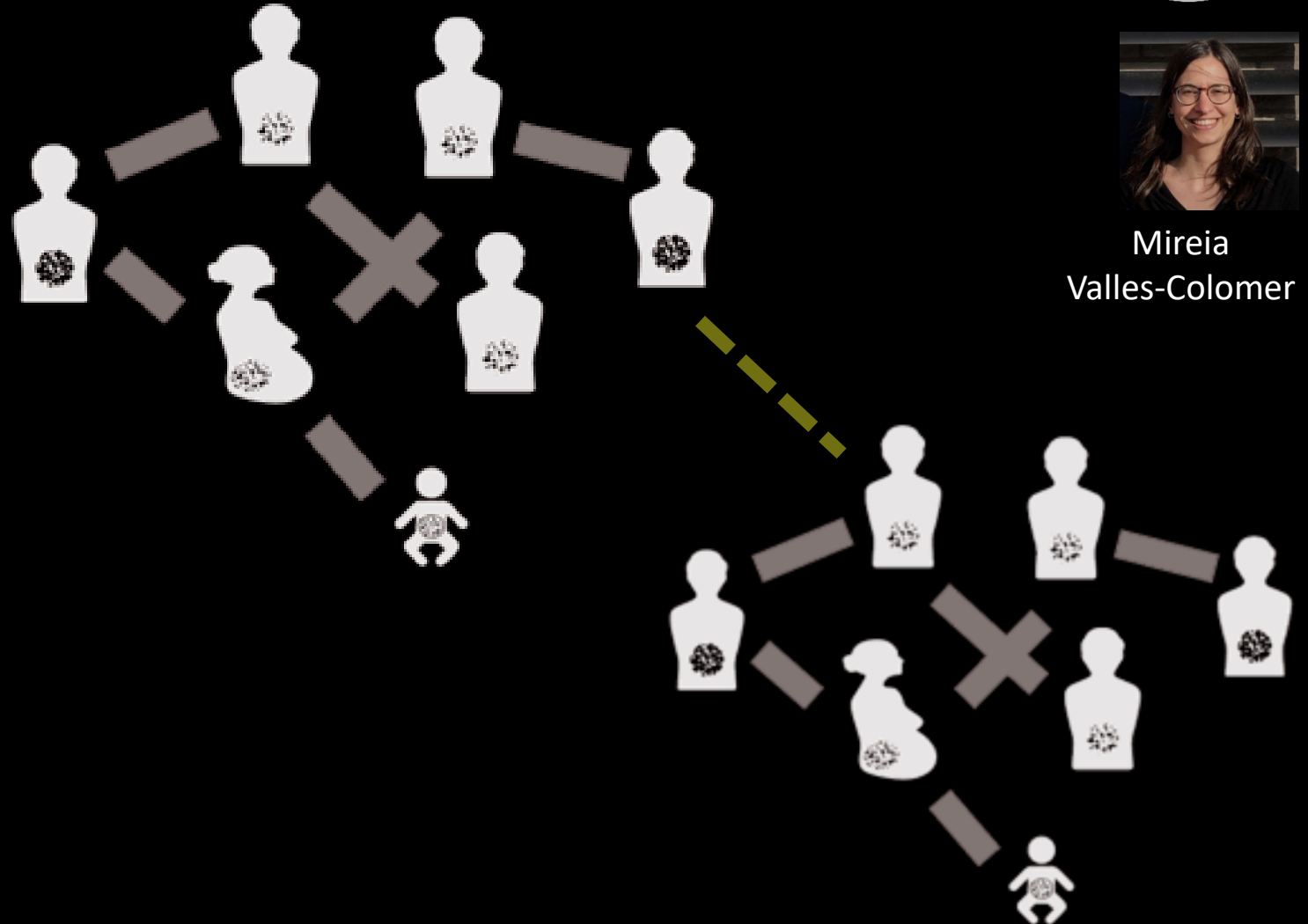
Mireia Valles-Colomer

# On the transmission of the human microbiome



Microbiome transmission set (N=9,715 [2,880])

Gut	Longitudinal sets	Mother-offspring pairs
	1,676 samples [0] 495 individuals [0] 5 datasets [0] 4 countries [0]	3,598 samples [636] 2,144 individuals [593] 711 pairs [261] 18 datasets [8] 14 countries [3]
Oral	Households	Adult twins
	1,123 samples [435] 952 individuals [435] 211 households [70] 7 datasets [3] 8 countries [1]	1,734 samples [0] 1,734 individuals [0] 712 pairs [0] 3 datasets [0] 1 country [0]
Oral	Mother-offspring pairs	Households
	1,315 samples [1,286] 1,315 individuals [1,286] 668 pairs [653] 2 datasets [1] 2 countries [1]	2,069 samples [1,929] 2,069 individuals [1,929] 730 households [646] 2 datasets [1] 2 countries [1]

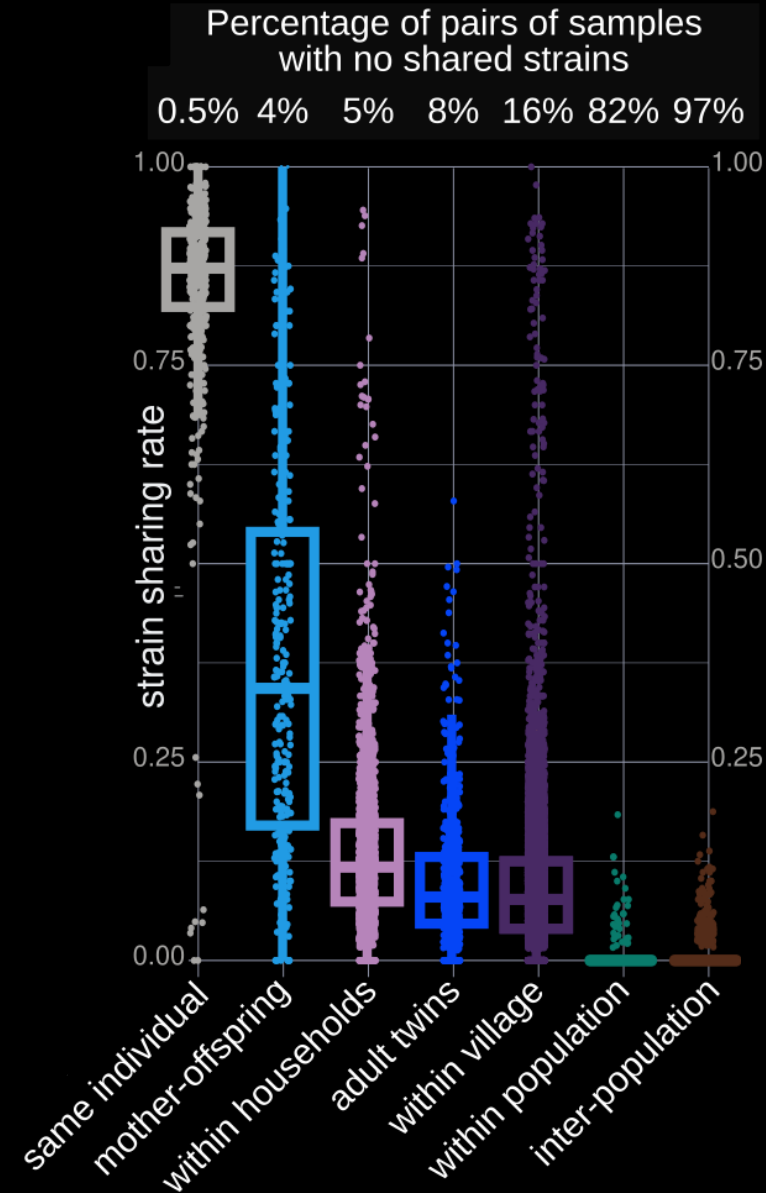
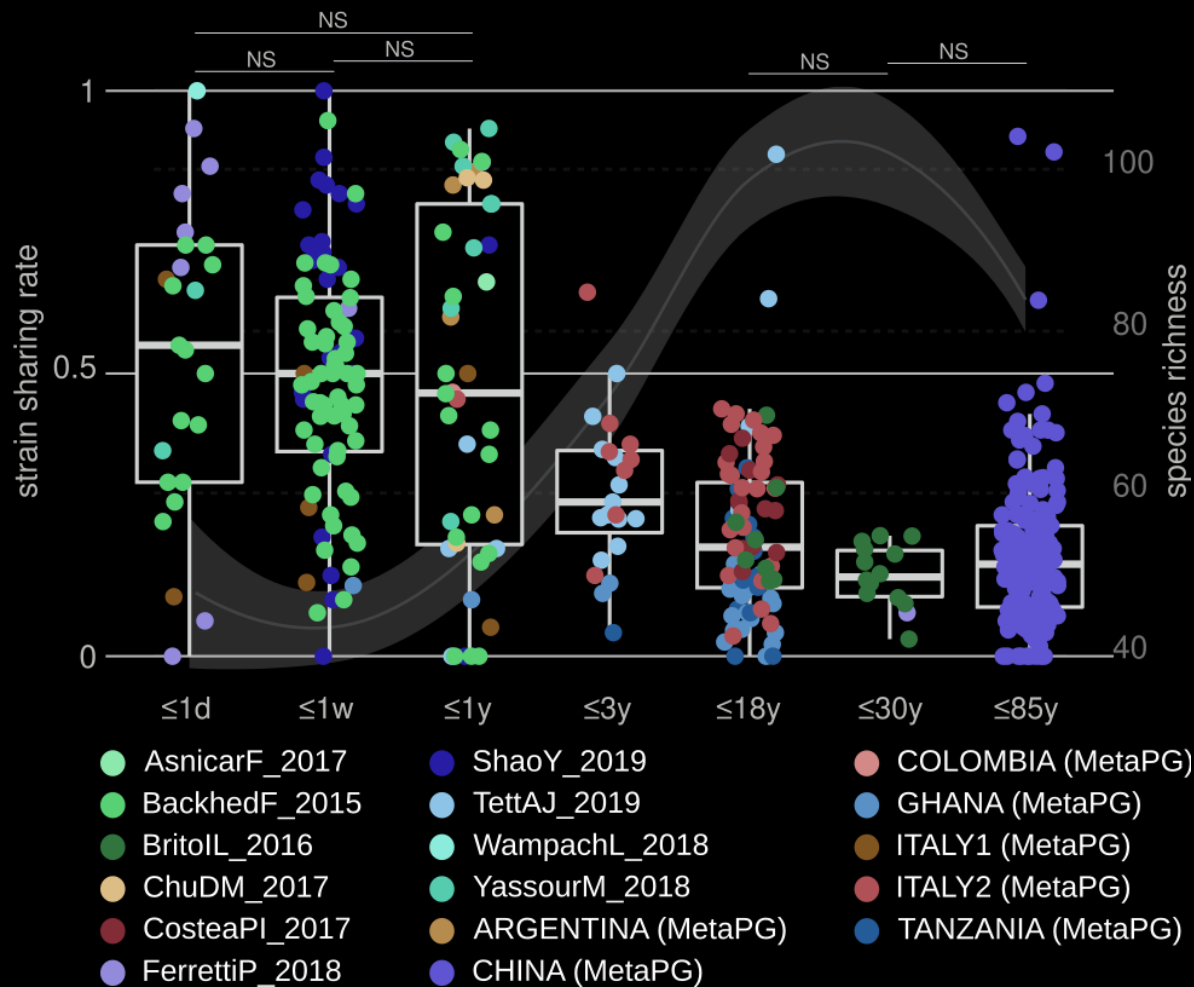
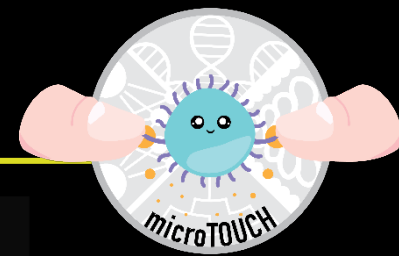


Mireia Valles-Colomer

Strain-level metagenomic profiling (StrainPhlAn)

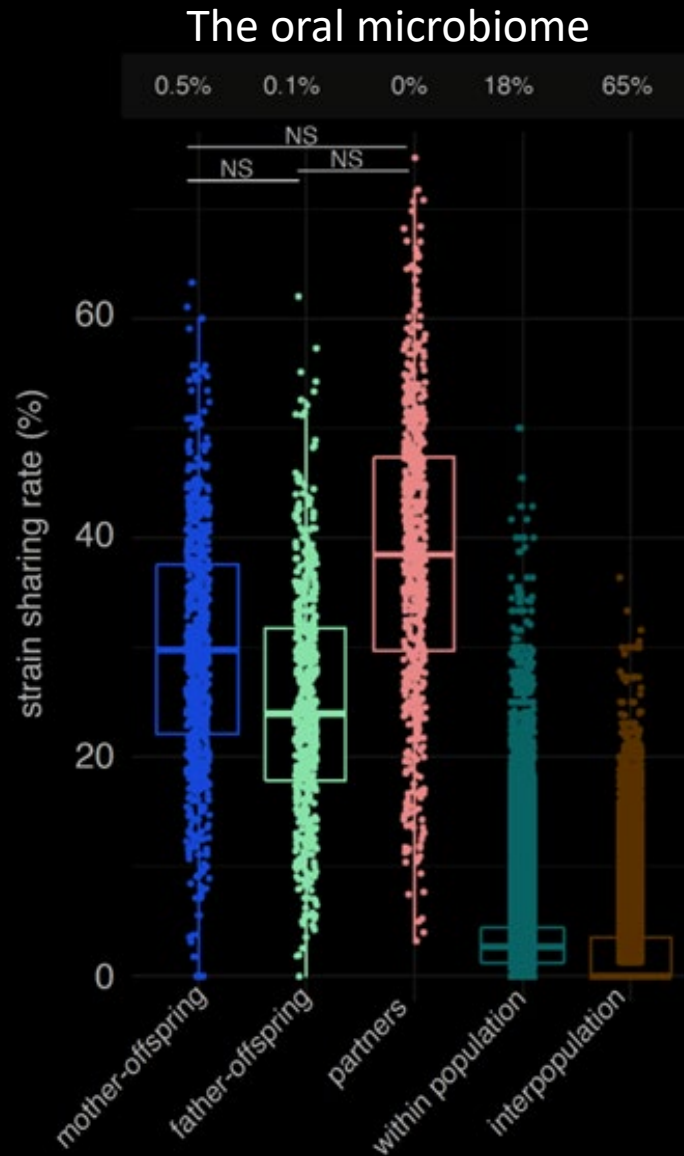
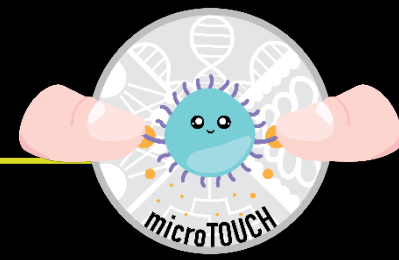
Gut	646 SGBs (365 uSGBs) ➤	477,141 strains
Oral	252 SGBs (60 uSGBs) ➤	326,403 strains

# On the transmission of the human microbiome



Mireia Valles-Colomer

# On the transmission of the human microbiome



**HYPOTHESIS**

**Science**

## Are noncommunicable diseases communicable?

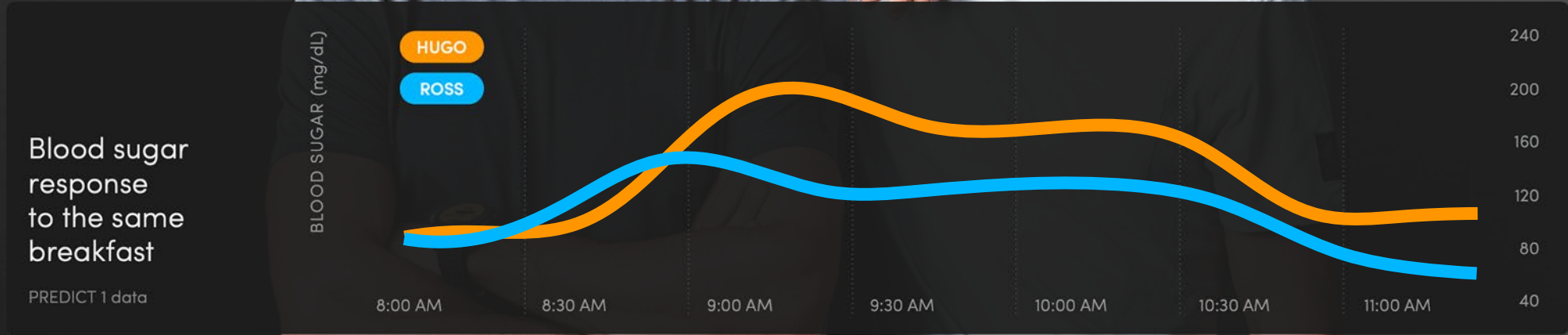
Numerous noncommunicable diseases could have a transmissible microbial component

By **B. B Finlay**<sup>1,2</sup> and **CIFAR Humans**



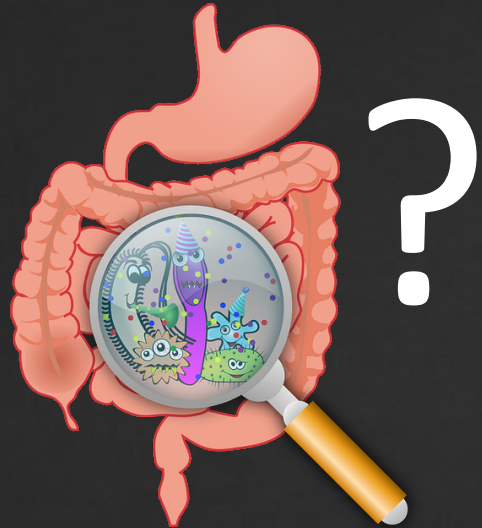
# We are all unique

With Tim Spector, Sarah Berry, TwinsUK, ZOE

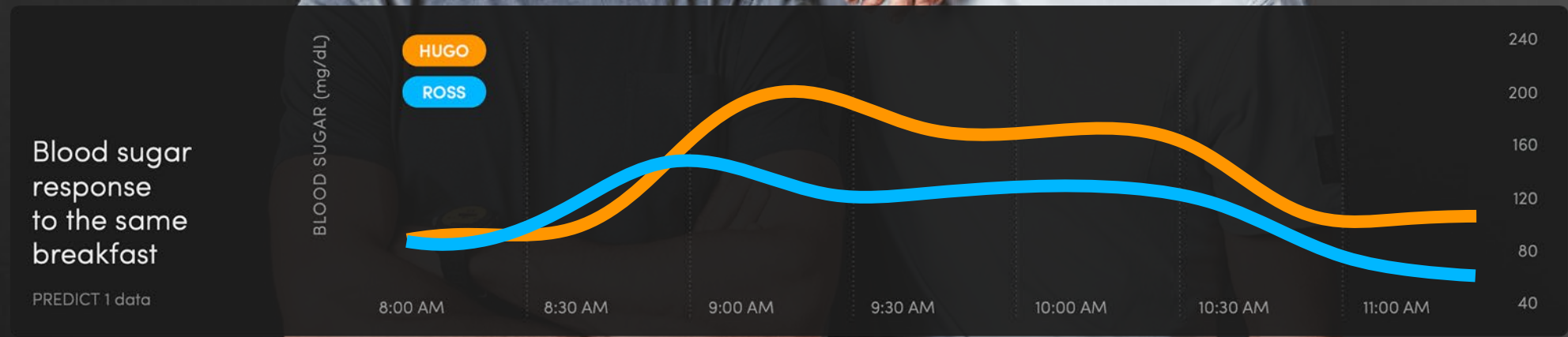
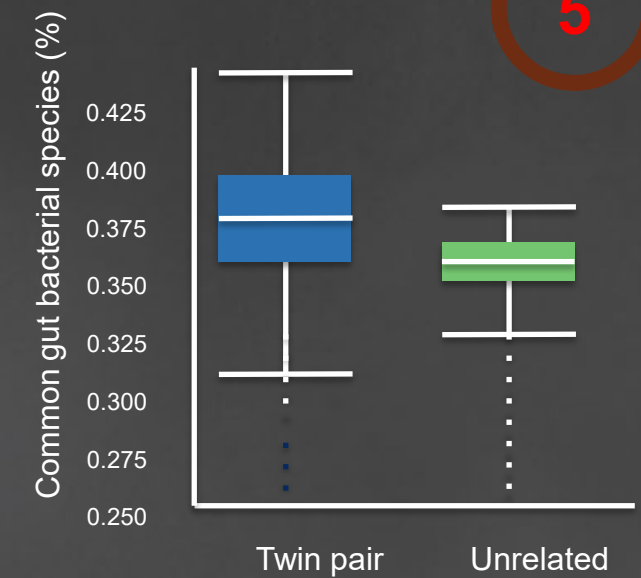


# We are all unique

...but the human microbiome is even more unique than us...  
...and could explain a lot of our phenotypic uniqueness!



5



# The PREDICT 1 Cohort

---

PREDICT 1 UK cohort



Discovery  
1,002 individuals

PREDICT 1 US cohort



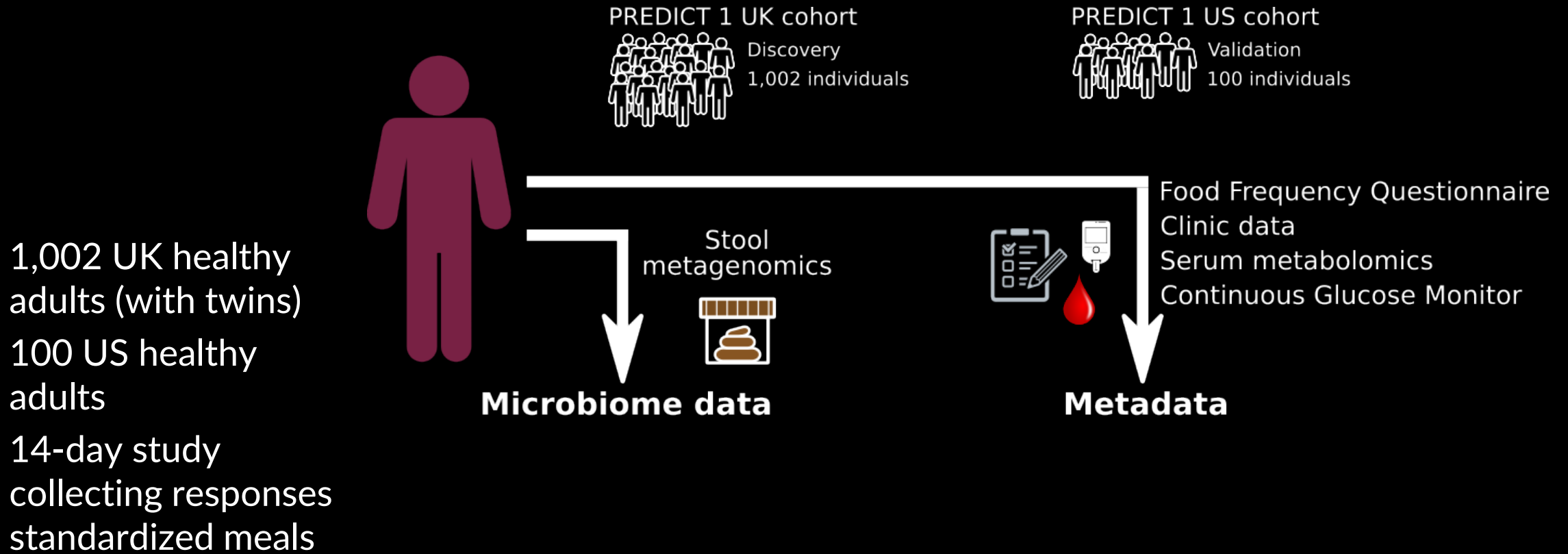
Validation  
100 individuals

1,002 UK healthy  
adults (with twins)

100 US healthy  
adults

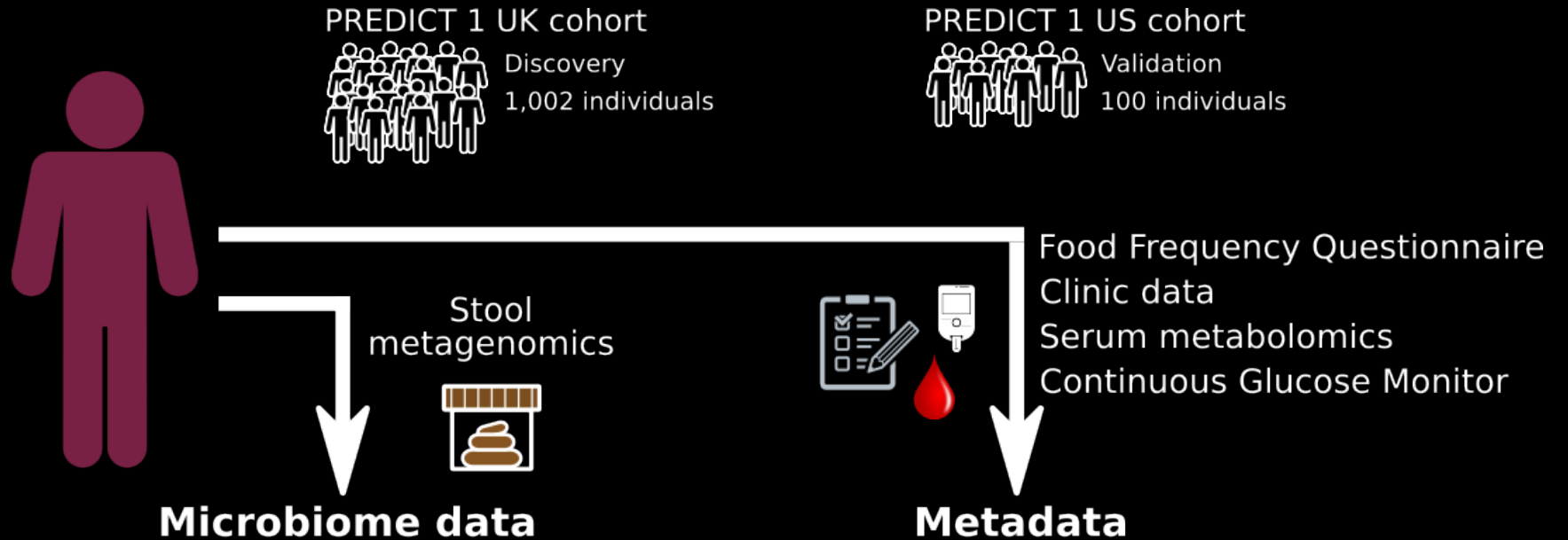
14-day study  
collecting responses  
standardized meals

# The PREDICT 1 Cohort



# The PREDICT 1 Cohort

1,002 UK healthy adults (with twins)  
 100 US healthy adults  
 14-day study  
 collecting responses  
 standardized meals



8.8 avg. 2.2 sd. Gb/sample 58.3 avg 14.6 sd. Mreads/sample			<b>1 Personal</b> Age, BMI, Weight, Waist/Hip ratio, Visceral fat, Antibiotic usage, Blood pressure Tot. 17	<b>2 Habitual diet</b> Foods, Food groups, Nutrients, Nutrients %E, Dietary patterns Tot. 223
Taxonomic	Functional	Assembly		
746 species	UniRef90 1,910,069	48,181 MAGs	<b>3 Fasting</b> Lipoproteins, ApoLipoproteins, Risk scores, Glucose mediated, FAs metabolism Tot. 269	<b>4 Post-prandial</b> All fasting measures up to 7 timepoints including max values and rises Tot. 1288
29 spp. 90% prevalent	UniRef50 878,520	29,035 MQ		
95 spp. 50% prevalent	KEGG KOs 6,163	19,146 HQ		
176 spp. 20% prevalent	Pathways 445			

# The PREDICT 1 Cohort

PREDICT 1 UK cohort



PREDICT 1 US cohort



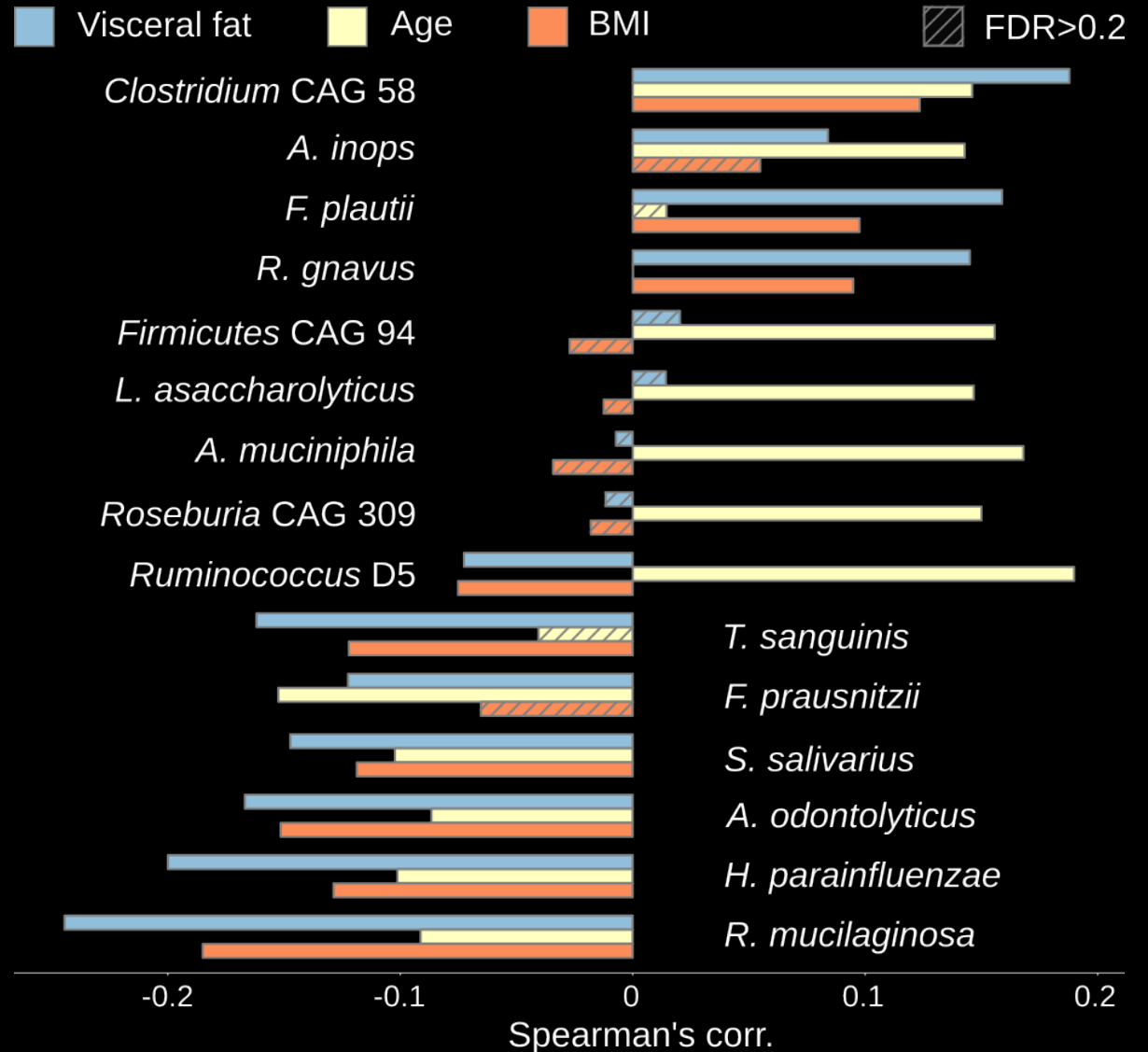
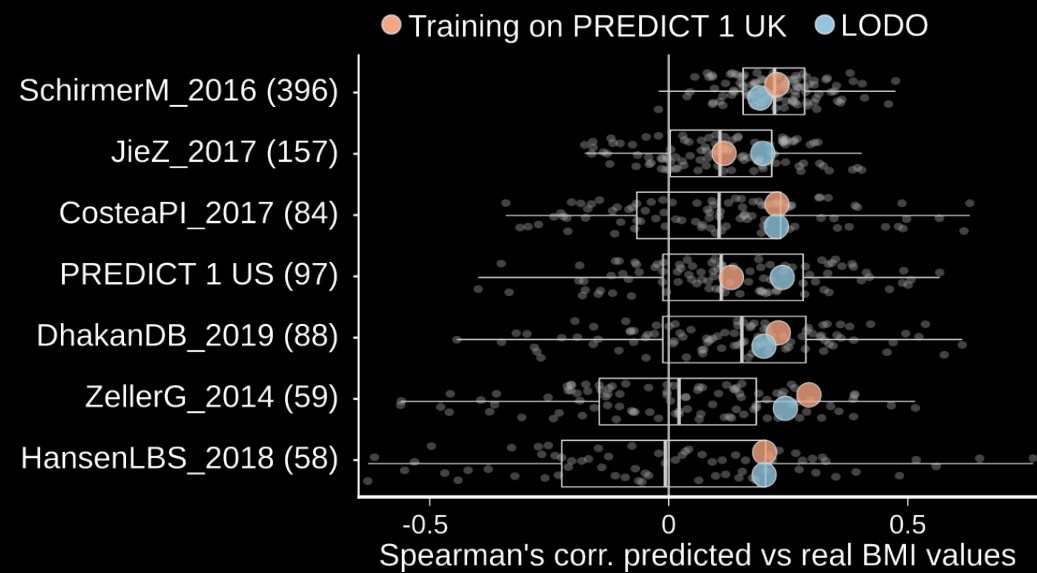
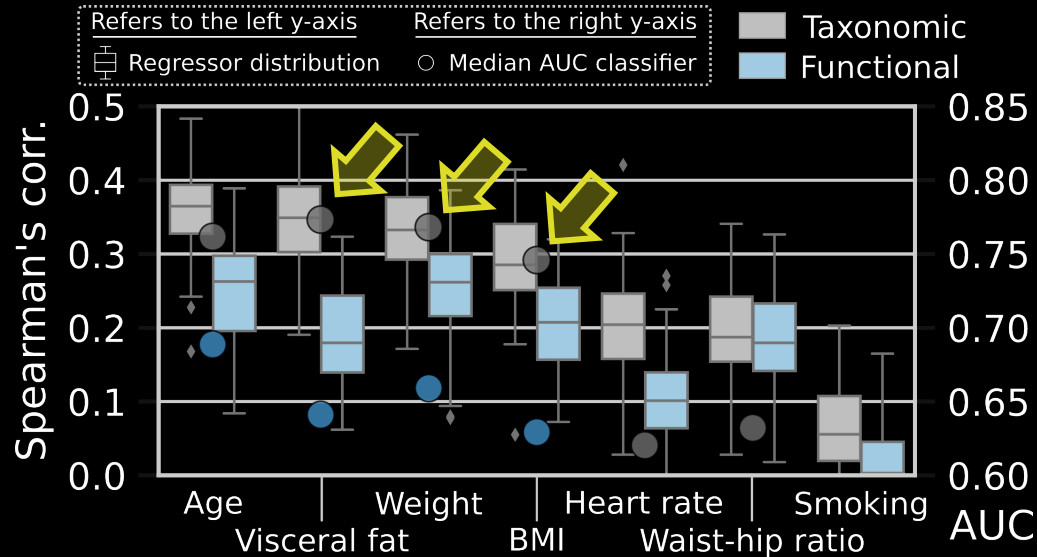
1,002 UK healthy adults (with twins)  
100 US healthy adults  
14-day study collecting responses standardized meals



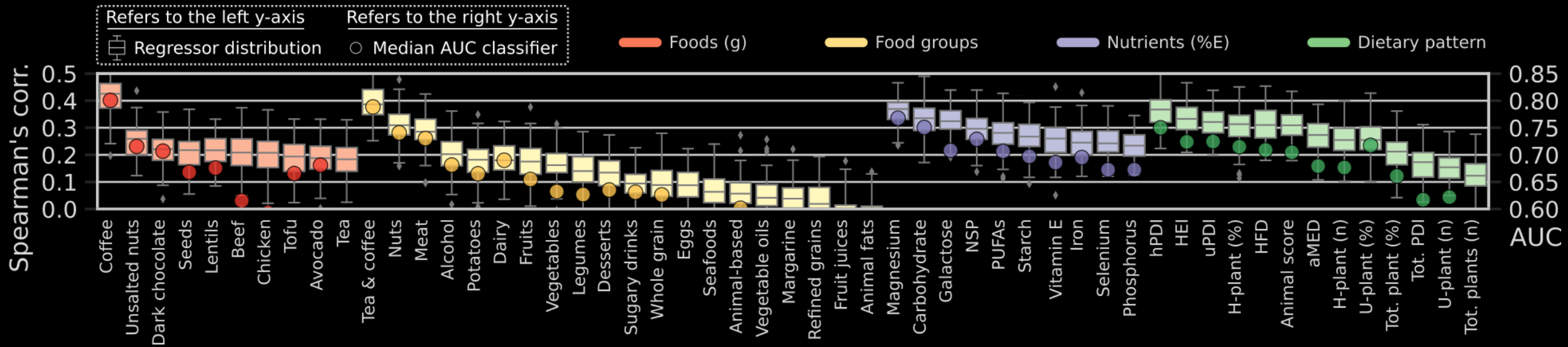
Food Frequency Questionnaire  
Clinic data  
Serum metabolomics  
Continuous Glucose Monitor

8.8 avg. 2.2 sd. Gb/sample 58.3 avg 14.6 sd. Mreads/sample			<b>1 Personal</b> Age, BMI, Weight, Waist/Hip ratio, Visceral fat, Antibiotic usage, Blood pressure Tot. 17	<b>2 Habitual diet</b> Foods, Food groups, Nutrients, Nutrients %E, Dietary patterns Tot. 223
<b>Taxonomic</b>	<b>Functional</b>	<b>Assembly</b>		
746 species	UniRef90 1,910,069	48,181 MAGs	<b>3 Fasting</b> Lipoproteins, ApoLipoproteins, Risk scores, Glucose mediated, FAs metabolism Tot. 269	<b>4 Post-prandial</b> All fasting measures up to 7 timepoints including max values and rises Tot. 1288
29 spp. 90% prevalent	UniRef50 878,520	29,035 MQ		
95 spp. 50% prevalent	KEGG KOs 6,163	19,146 HQ		
176 spp. 20% prevalent	Pathways 445			

# PREDICT 1: strong and reproducible links with obesity markers



# PREDICT 1: strong links with foods and dietary patterns

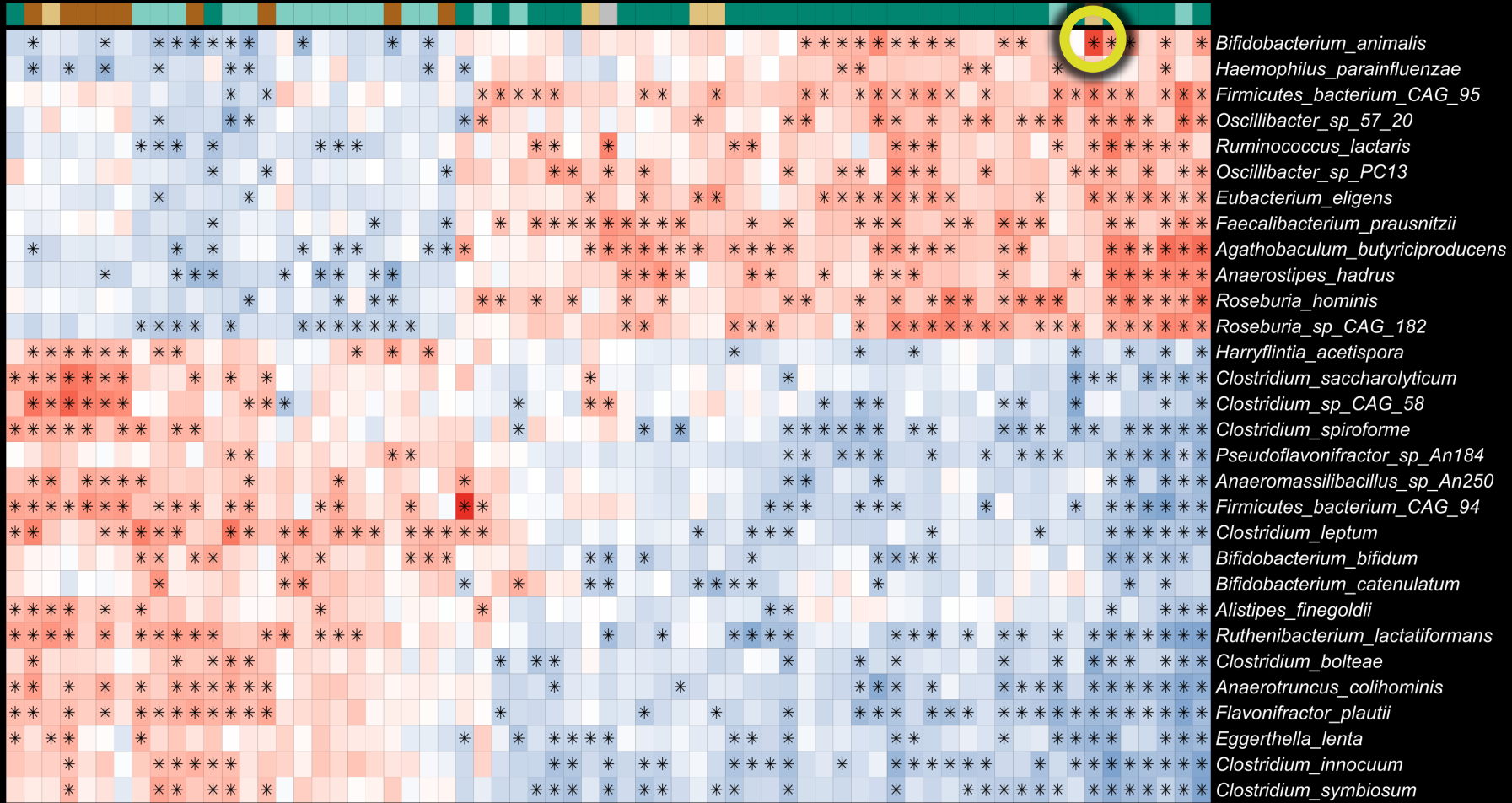


Nutritionists processed FFQs and derived single foods, food groups, nutrients, and dietary indexes



# PREDICT 1: a map of direct microbe-food associations

■ Healthy plant-based  
 ■ Less Healthy plant-based  
 ■ Healthy animal-based  
 ■ Unhealthy animal-based  
 ■ No category



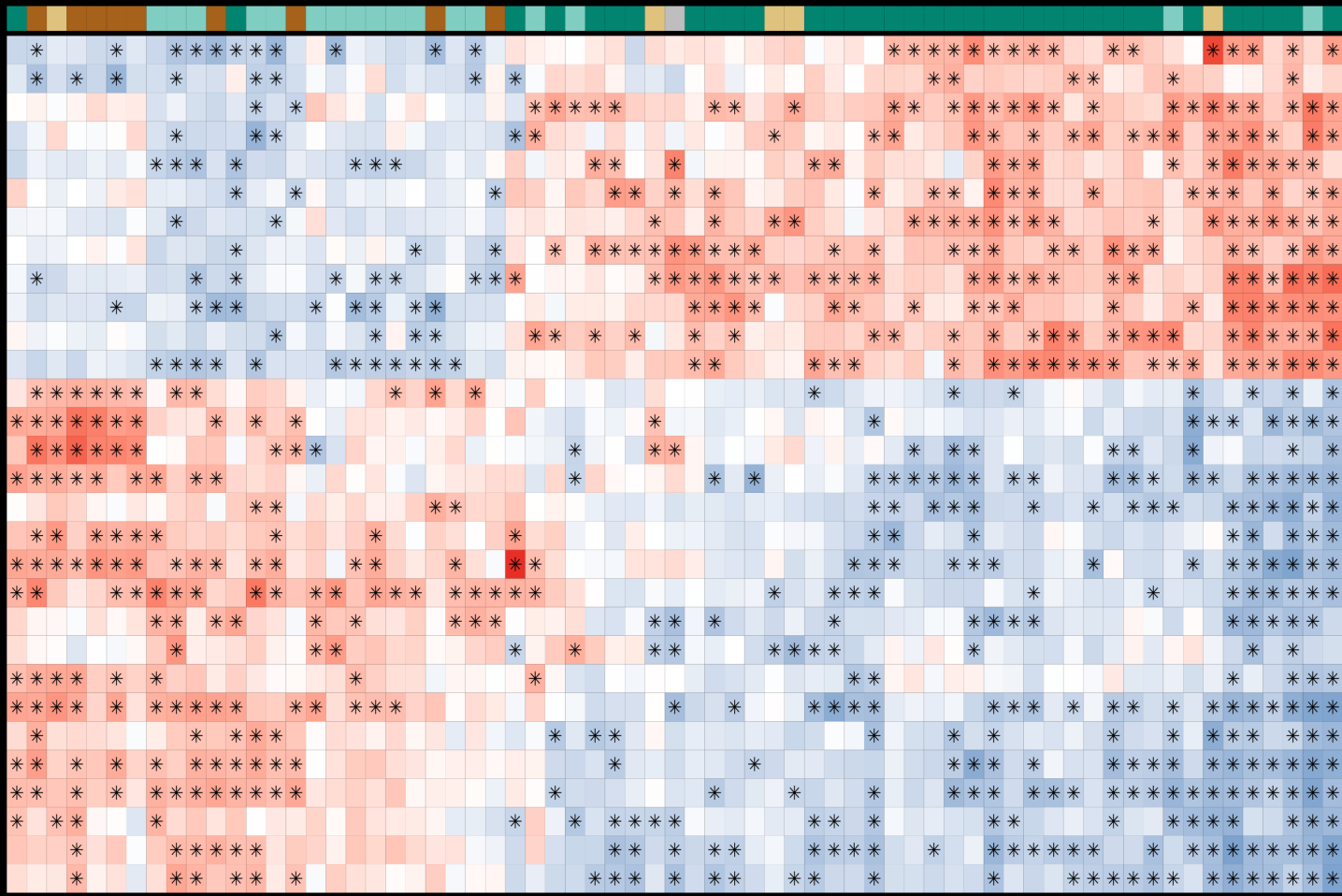
Sauces    Beans    Fruit juice    Coffee    Eggs    Tomatoes    Fullfat yogurt    Seeds  
 Bacon    Savoury pies    Dairy dessert    Spinach

Spearman corr.

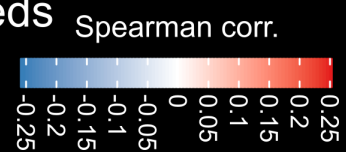
-0.25   -0.15   -0.05   0   0.05   0.15   0.25

# PREDICT 1: a map of microbe-food associations

■ Healthy plant-based  
 ■ Less Healthy plant-based  
 ■ Healthy animal-based  
 ■ Unhealthy animal-based  
 ■ No category

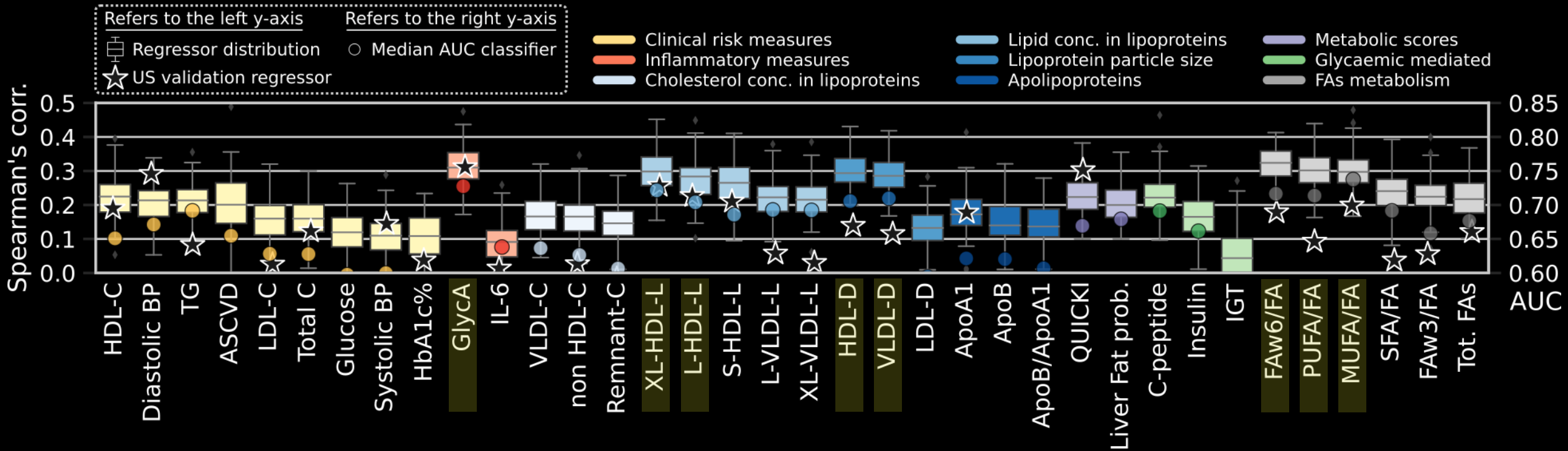


Sauces    Beans    Fruit juice    Coffee    Eggs    Tomatoes    Fullfat yogurt    Seeds  
 Bacon    Savoury pies    Dairy dessert    Spinach



- Diets rich in **“healthy”, plant-based foods** associated with **“good”** gut microbes
- “Unhealthy”** microbe cluster was seen in those eating **“unhealthy” plant and “unhealthy” animal-based foods**
- Highlights the importance of food quality
- Overall dietary patterns matter

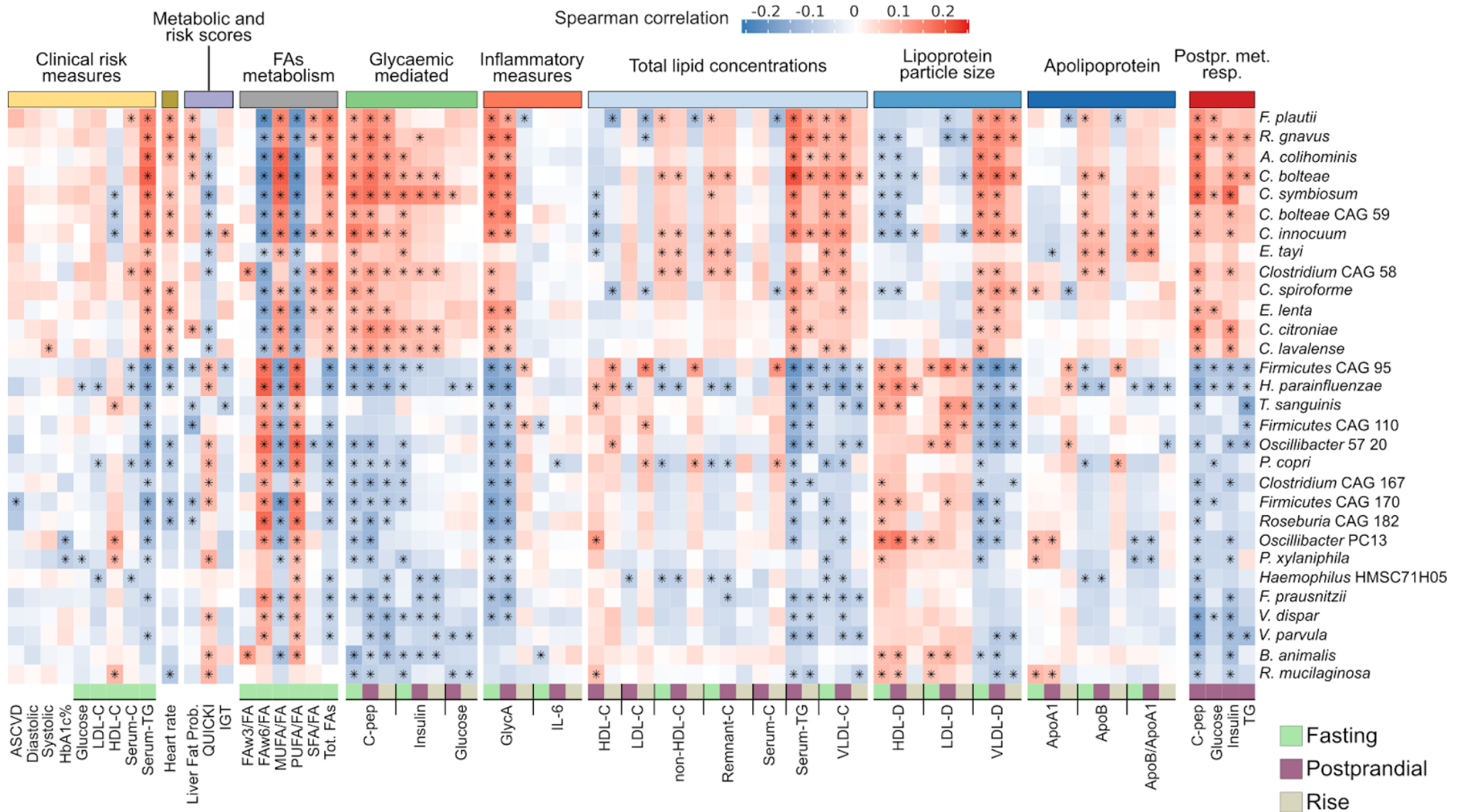
# PREDICT 1: strong links with metabolic markers



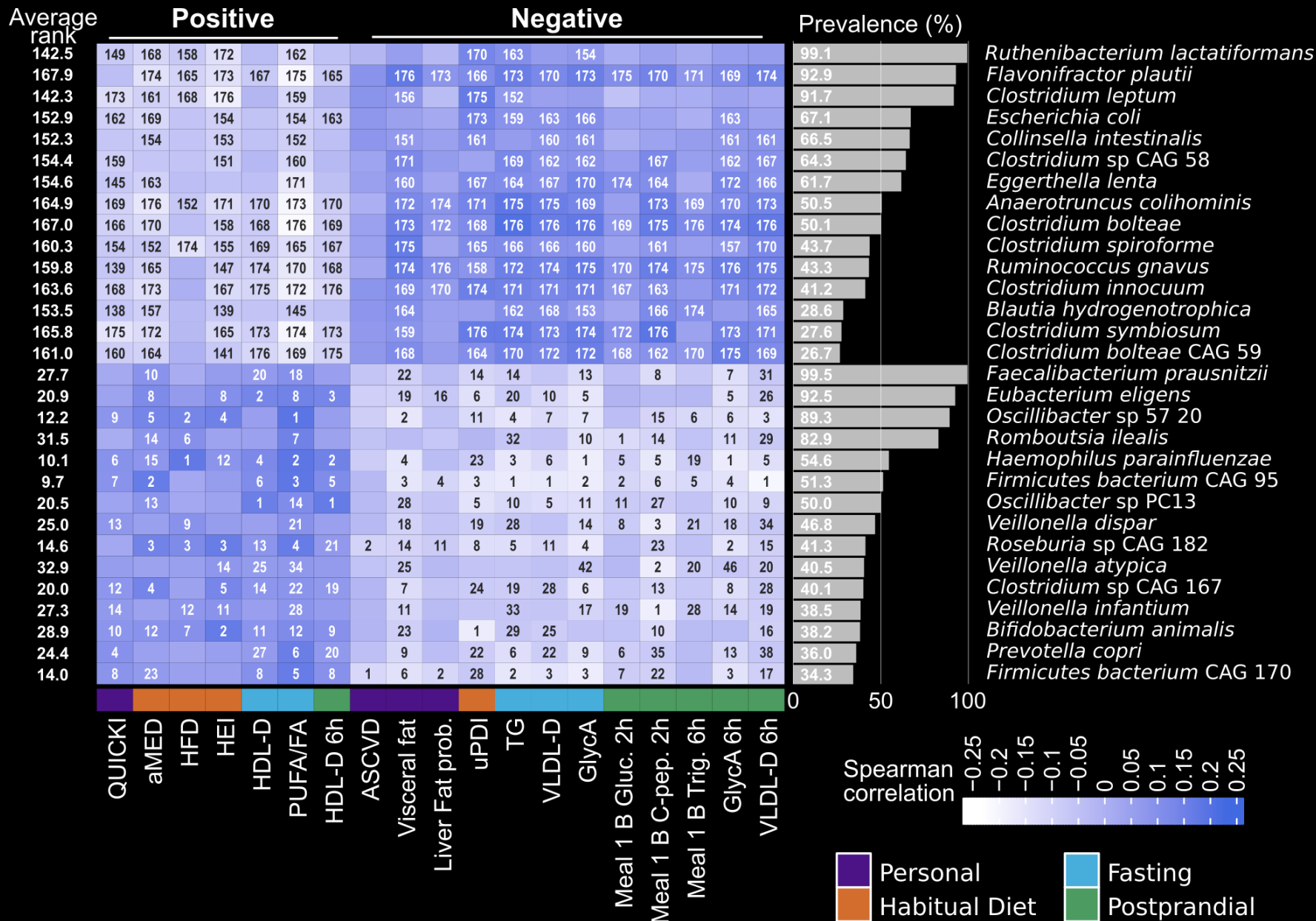
Clinical risk measures not strongly linked with the microbiome

GlycA and several cholesterol and fatty acids markers show a strong link with the microbiome

# But the scenario is more complex when considering postprandial responses...



# PREDICT 1: an overall signature of favourable/unfavourable microbes



# Better characterization of the gut microbiome w.r.t. transit time: the blue poo



Asnicar *et al*, Gut 2021



## Questionnaire

Diet (FFQ)  
Stool form and frequency



## Microbiome analysis

Stool sample



## Blood samples

Fasting and post-prandial measures



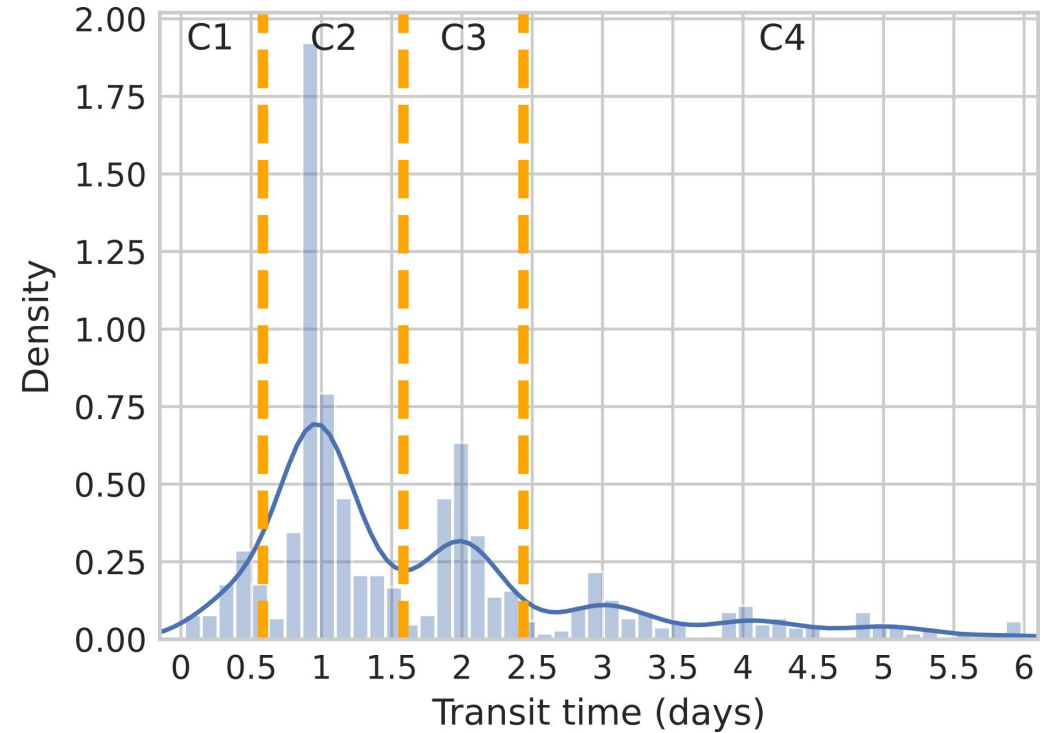
## Transit time (hrs)



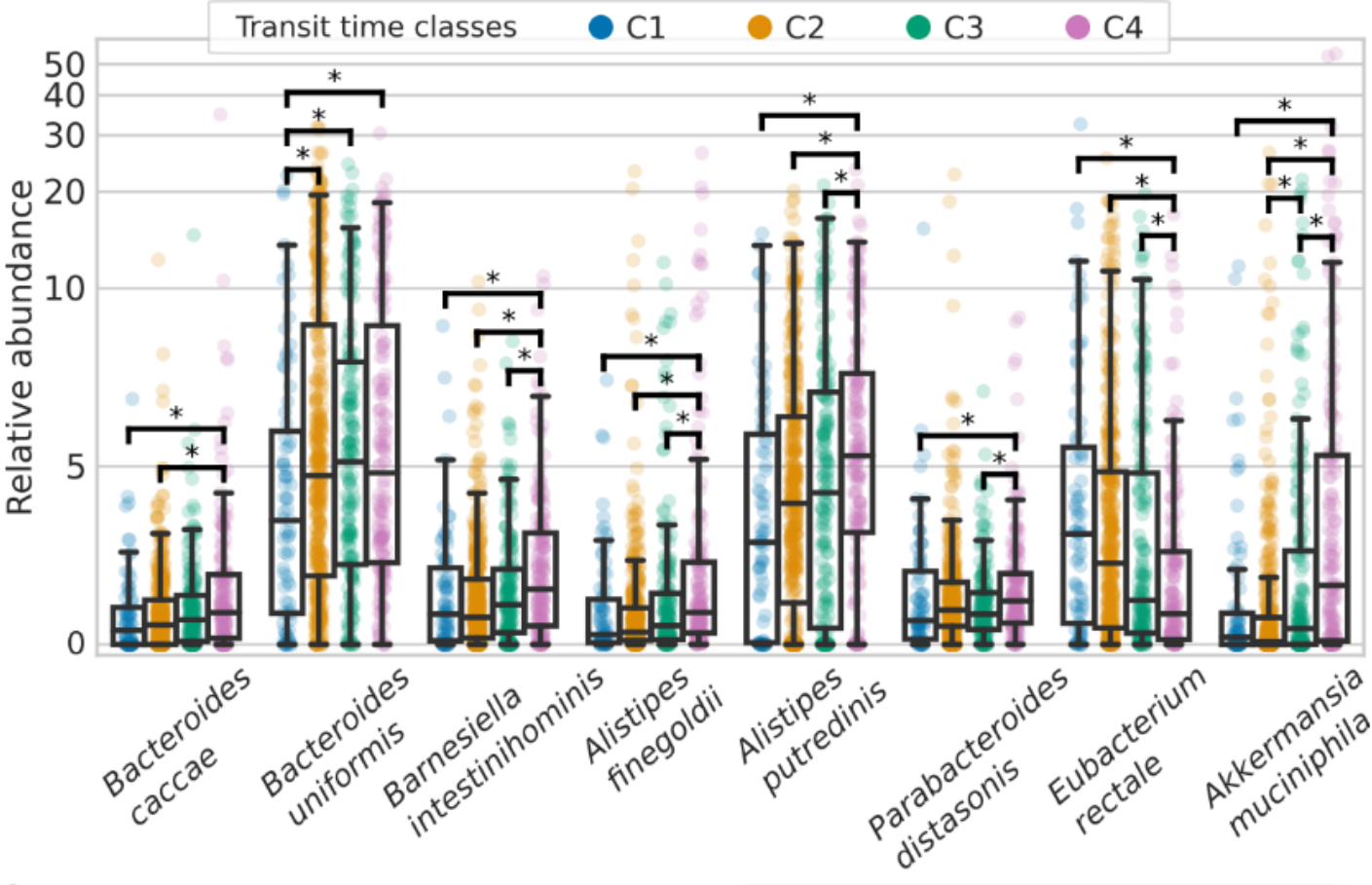
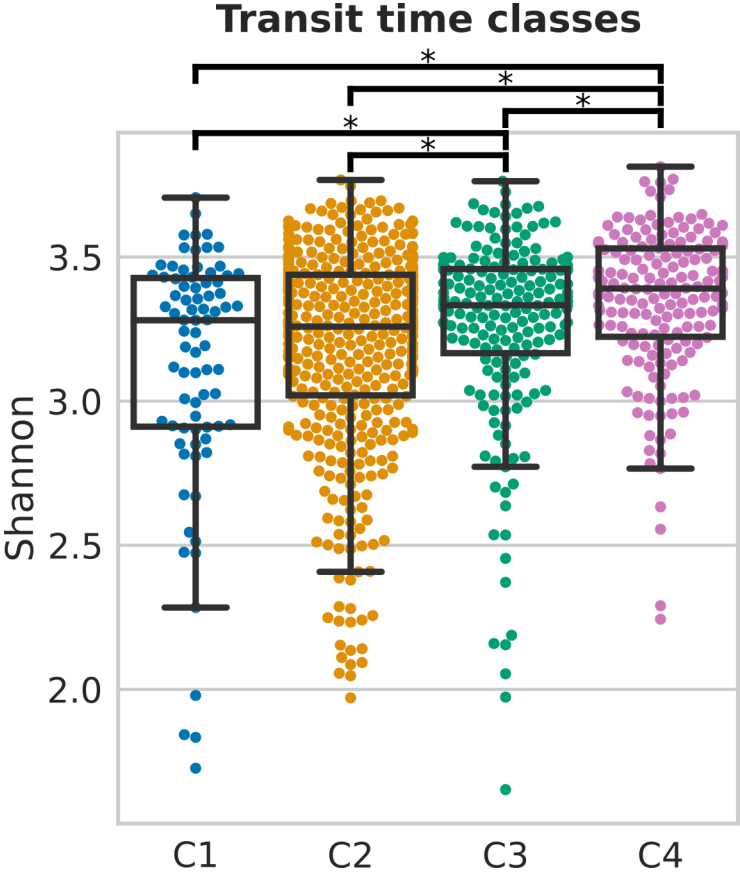
Within 24hr prior to clinic visit

Clinic Visit

Self-report on mobile app



# Better characterization of the gut microbiome w.r.t. transit time: the blue poo



# How can we harness our findings to achieve meaningful change?

Machine learning algorithms allow us to predict how each individual may respond to foods.

This enables us to make personalized dietary recommendations at the individual level.





## Microbiome connections with host metabolism and habitual diet from 1,098 deeply phenotyped individuals

Francesco Asnicar<sup>1,16</sup>, Sarah E. Berry<sup>2,16</sup>, Ana M. Valdes<sup>3,4</sup>, Long H. Nguyen<sup>5</sup>, Gianmarco Piccinno<sup>1</sup>, David A. Drew<sup>5</sup>, Emily Leeming<sup>6</sup>, Rachel Gibson<sup>2</sup>, Caroline Le Roy<sup>6</sup>, Haya Al Khatib<sup>7</sup>, Lucy Francis<sup>7</sup>, Mohsen Mazidi<sup>6</sup>, Olatz Mompeo<sup>6</sup>, Mireia Valles-Colomer<sup>1</sup>, Adrian Tett<sup>1</sup>, Francesco Beghini<sup>1</sup>, Léonard Dubois<sup>1</sup>, Davide Bazzani<sup>1</sup>, Andrew Maltez Thomas<sup>1</sup>, Chloe Mirzayi<sup>8</sup>, Asya Khleborodova<sup>8</sup>, Sehyun Oh<sup>8</sup>, Rachel Hine<sup>7</sup>, Christopher Bonnett<sup>7</sup>, Joan Capdevila<sup>7</sup>, Serge Danzanvilliers<sup>7</sup>, Francesca Giordano<sup>7</sup>, Ludwig Geistlinger<sup>8</sup>, Levi Waldron<sup>8</sup>, Richard Davies<sup>7</sup>, George Hadjigeorgiou<sup>7</sup>, Jonathan Wolf<sup>7</sup>, José M. Ordovás<sup>9,10</sup>, Christopher Gardner<sup>11</sup>, Paul W. Franks<sup>12,13</sup>, Andrew T. Chan<sup>10,5,13,14,17</sup>, Curtis Huttenhower<sup>13,14,17</sup>, Tim D. Spector<sup>6,17</sup> and Nicola Segata<sup>1,15,17</sup>

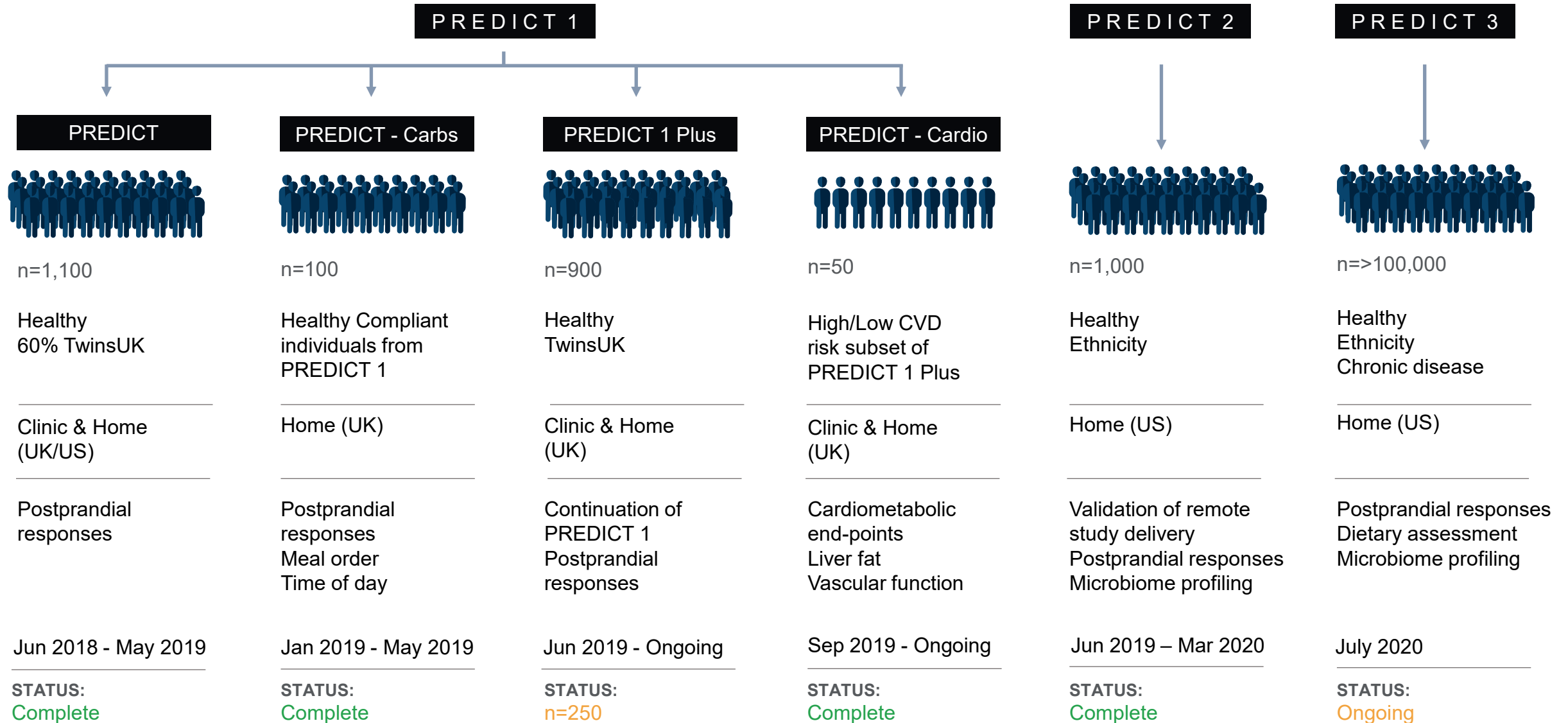
## Human postprandial responses to food and potential for precision nutrition

Sarah E. Berry<sup>1,18</sup>, Ana M. Valdes<sup>2,3,18</sup>, David A. Drew<sup>4</sup>, Francesco Asnicar<sup>5</sup>, Mohsen Mazidi<sup>6</sup>, Jonathan Wolf<sup>7</sup>, Joan Capdevila<sup>7</sup>, George Hadjigeorgiou<sup>7</sup>, Richard Davies<sup>7</sup>, Haya Al Khatib<sup>1,7</sup>, Christopher Bonnett<sup>7</sup>, Sajaysurya Ganesh<sup>7</sup>, Elco Bakker<sup>7</sup>, Deborah Hart<sup>6</sup>, Massimo Mangino<sup>6</sup>, Jordi Merino<sup>8,9,10,11</sup>, Inbar Linenberg<sup>7</sup>, Patrick Wyatt<sup>7</sup>, Jose M. Ordovas<sup>12,13</sup>, Christopher D. Gardner<sup>14</sup>, Linda M. Delahanty<sup>15</sup>, Andrew T. Chan<sup>4</sup>, Nicola Segata<sup>5,18</sup>, Paul W. Franks<sup>6,16,17,18</sup> and Tim D. Spector<sup>6,18</sup>

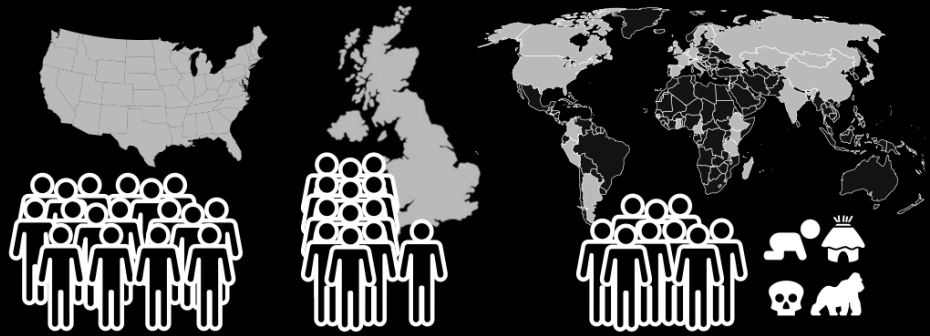
## Blue poo: impact of gut transit time on the gut microbiome using a novel marker

Francesco Asnicar,<sup>1</sup> Emily R Leeming<sup>2</sup>, Eirini Dimidi,<sup>3</sup> Mohsen Mazidi,<sup>2</sup> Paul W Franks,<sup>4</sup> Haya Al Khatib,<sup>3,5</sup> Ana M Valdes,<sup>6</sup> Richard Davies<sup>5</sup>, Elco Bakker,<sup>5</sup> Lucy Francis,<sup>5</sup> Andrew Chan,<sup>7</sup> Rachel Gibson,<sup>3</sup> George Hadjigeorgiou,<sup>5</sup> Jonathan Wolf,<sup>5</sup> Timothy D Spector,<sup>8</sup> Nicola Segata,<sup>9</sup> Sarah E Berry<sup>3</sup>

# Next steps on unravelling microbiome-diet links



# The microbiome and coffee on 30k individuals



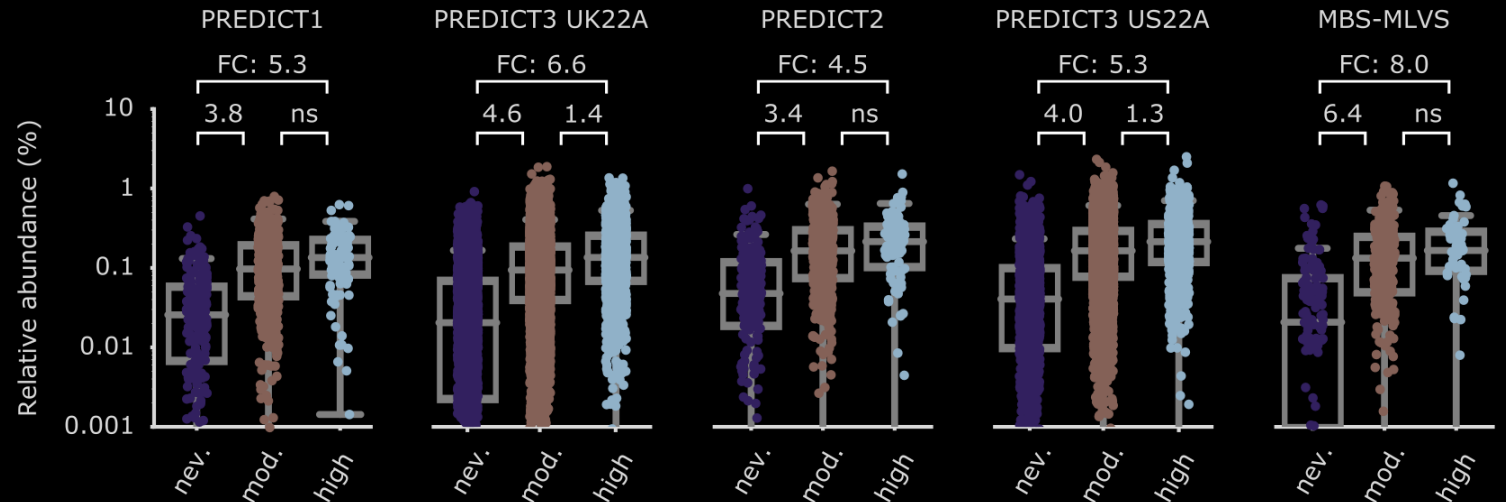
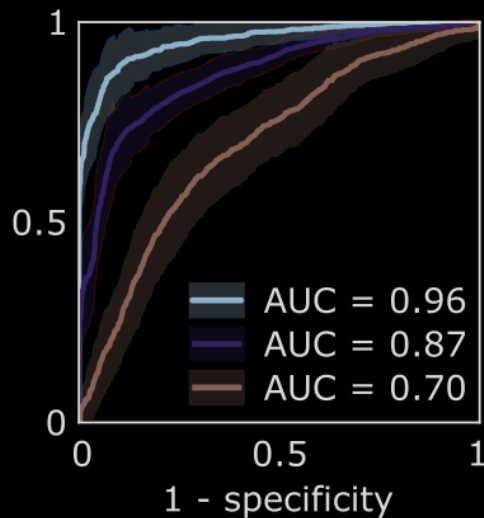
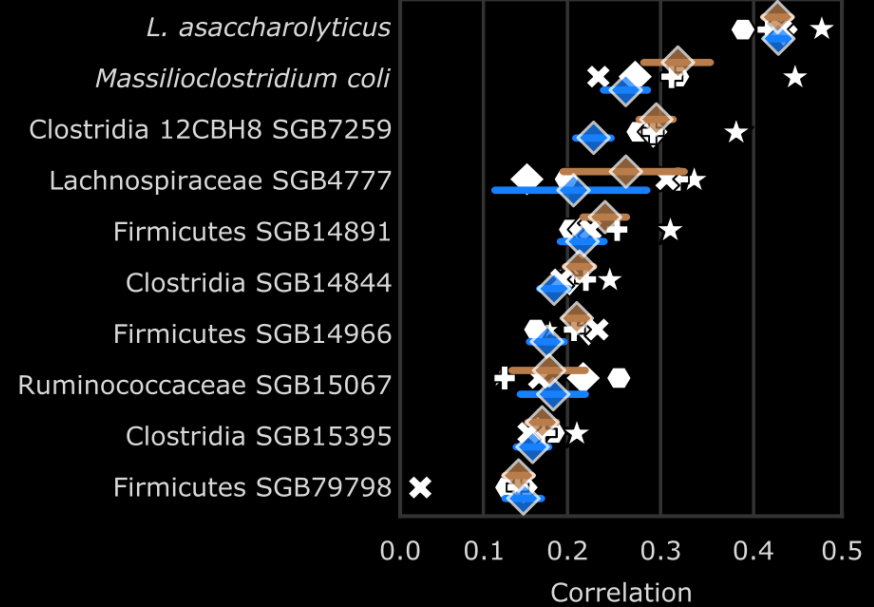
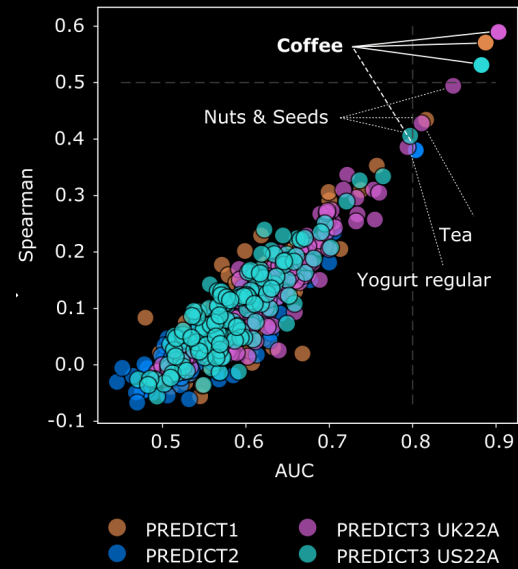
## The ZOE PREDICT cohorts:

Four US cohorts:  
 P2, n=975  
 P3 US21, n=11,798  
 P3 US22A, n=8,470

Two UK cohorts:  
 P1, n=1,098  
 P3 UK22A, n=12,353

## Public cohorts:

52 healthy studies, n=6,955  
 ten babies studies, n=977  
 20 non-west., n=1,413  
 NHP (n=203), ancient (n=29)

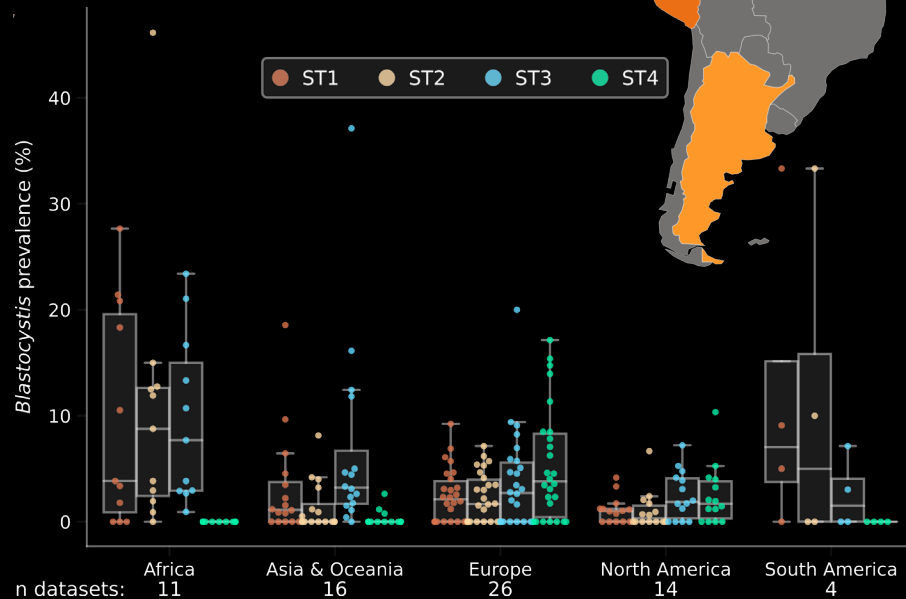
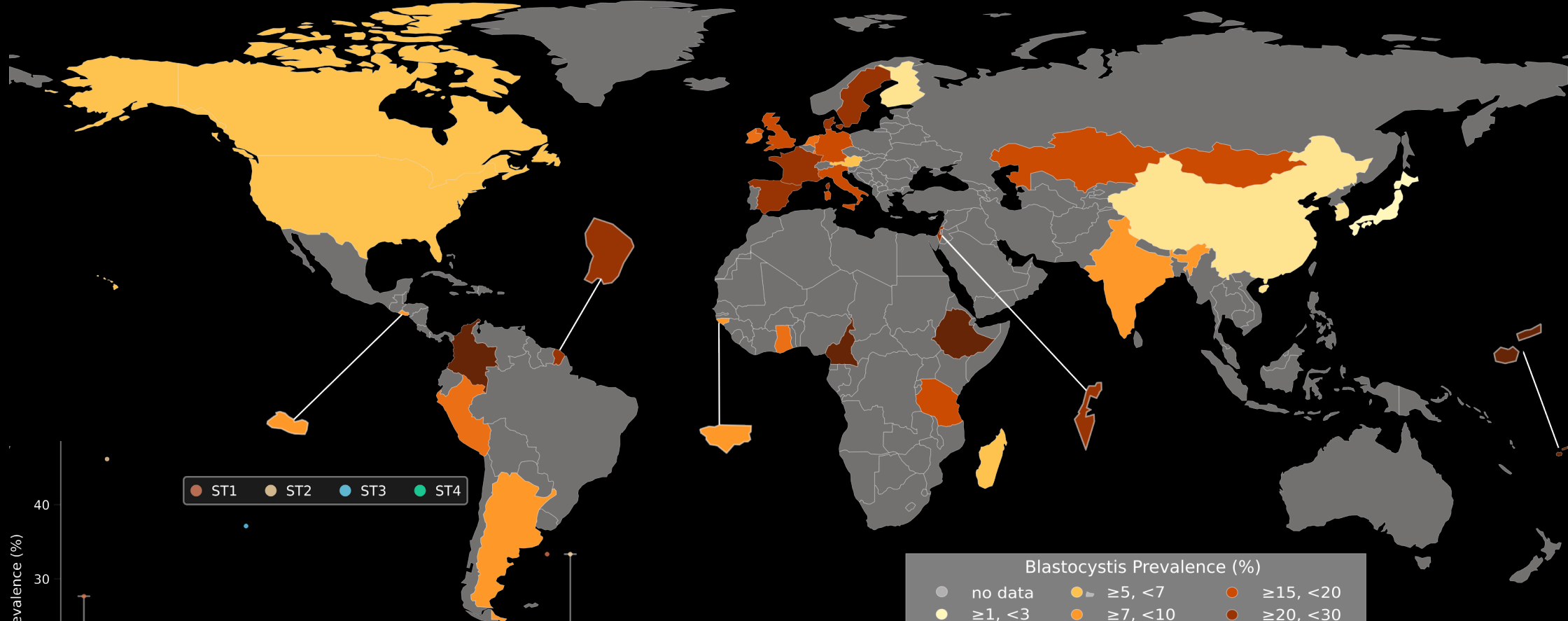


# Blastocystis is a prevalent “parasite”

8



Elisa Piperni

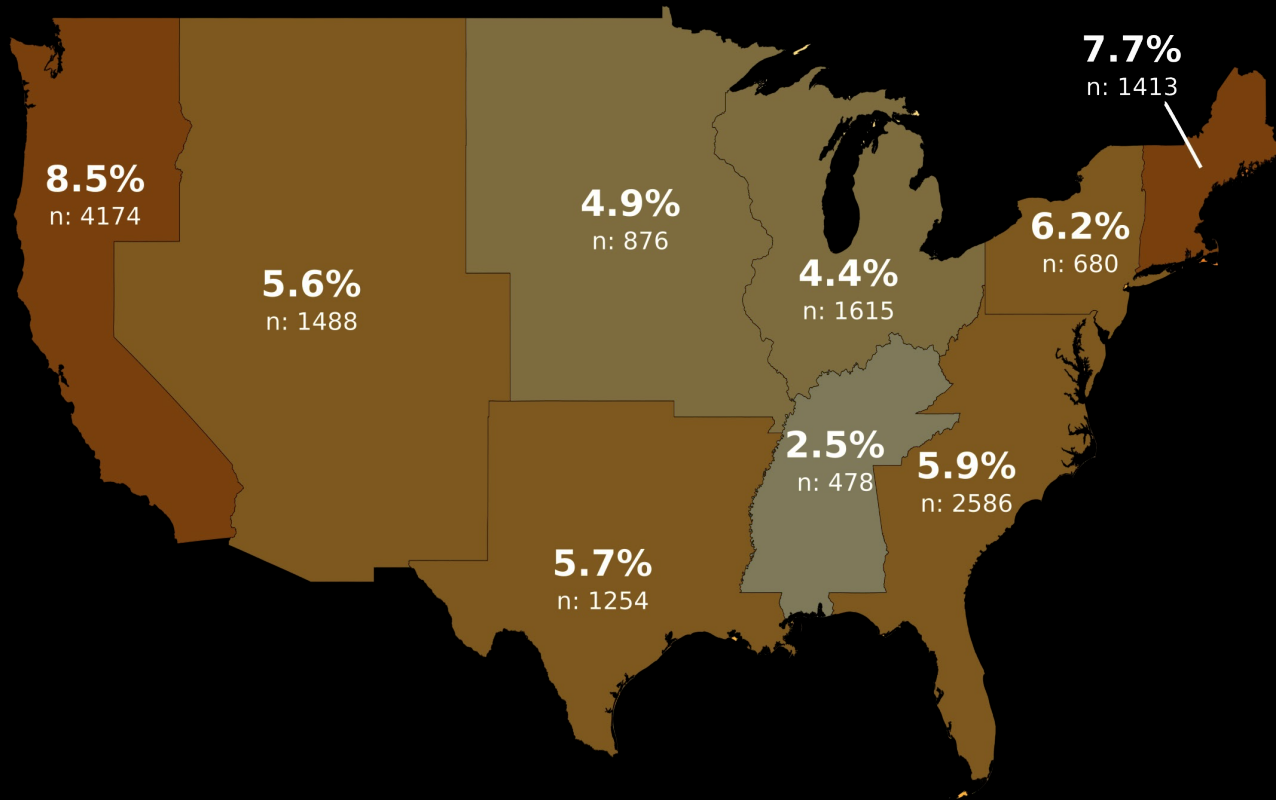


- Large differences in prevalence worldwide!
- ST3 specific to Westernized continents

# US has low prevalence of *Blastocystis*

---

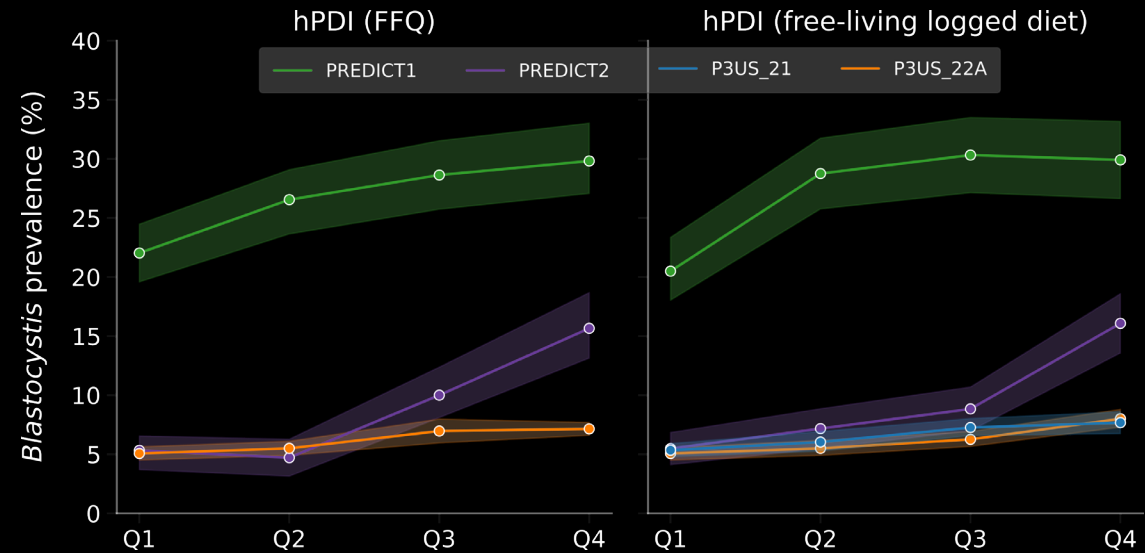
- On 14,564 healthy individuals from all over the US
- Higher prevalence on coastal regions than hinterlands





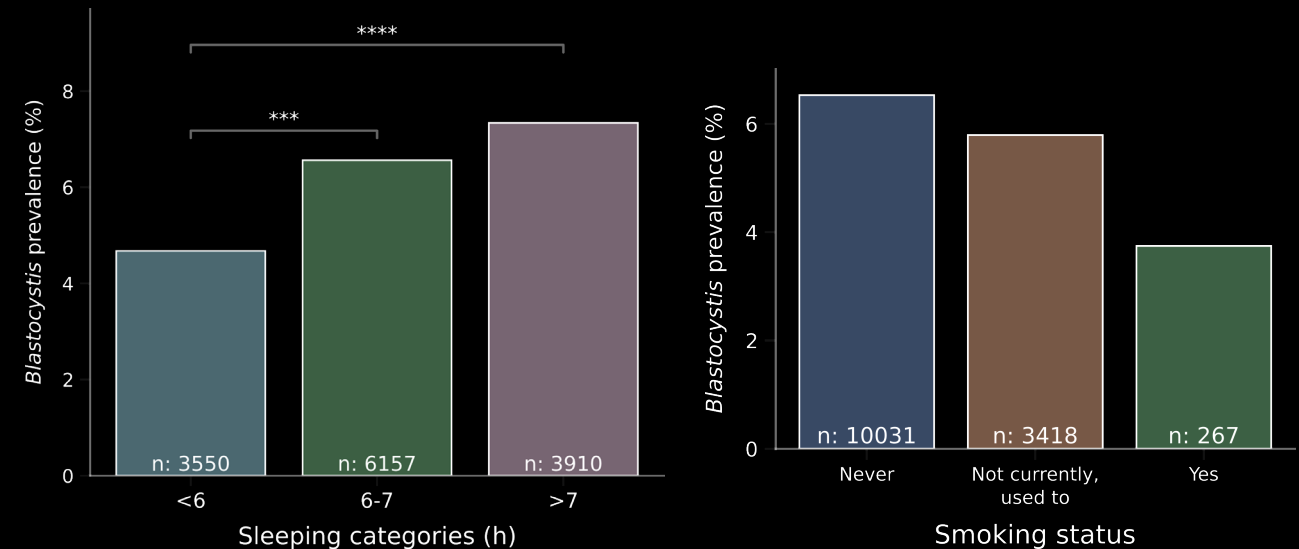
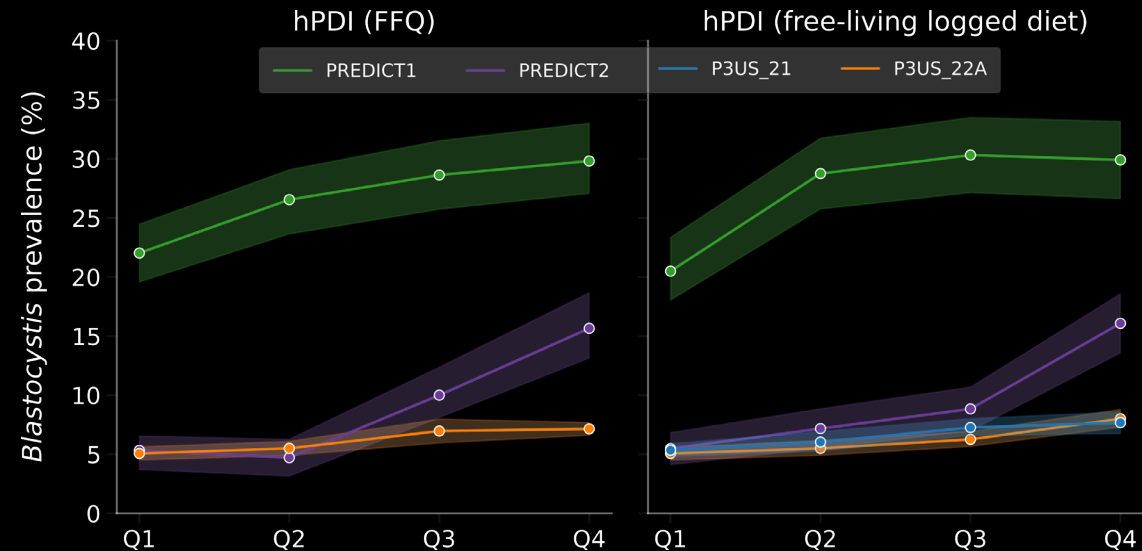
# *Blastocystis* associated with a healthier lifestyle

- We can assess 'diet quality' via dietary indices
- hPDI calculated from both FFQ and logged diet data



# Blastocystis associated with a healthier lifestyle

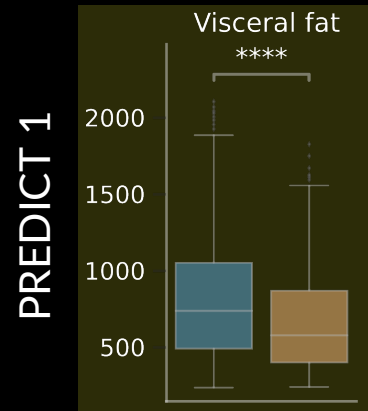
- We can assess 'diet quality' via dietary indices
- hPDI calculated from both FFQ and logged diet data
- Individuals with higher hPDI might be also more careful on sleep quality/quantity
- As well as to avoid smoking





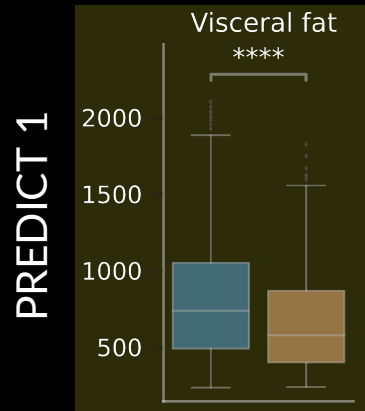
# *Blastocystis* strongly associated with low visceral fat

---

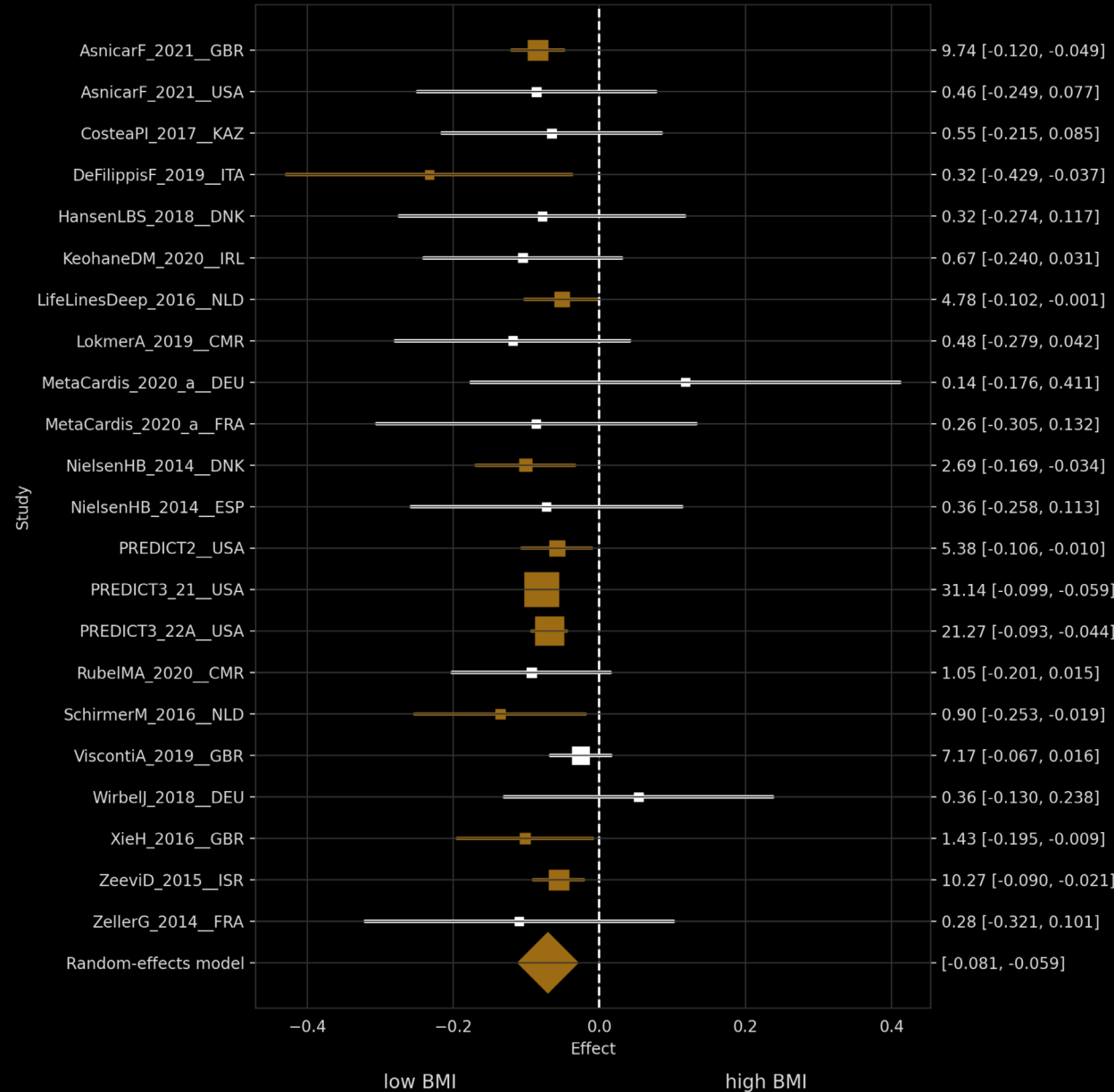
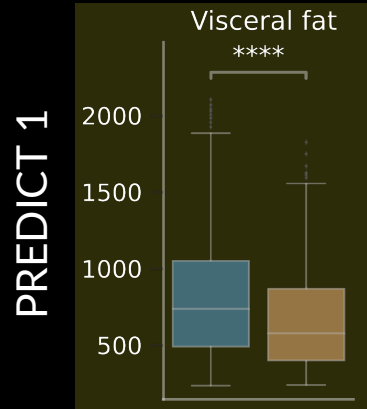


■ *Blastocystis* negative  
■ *Blastocystis* positive

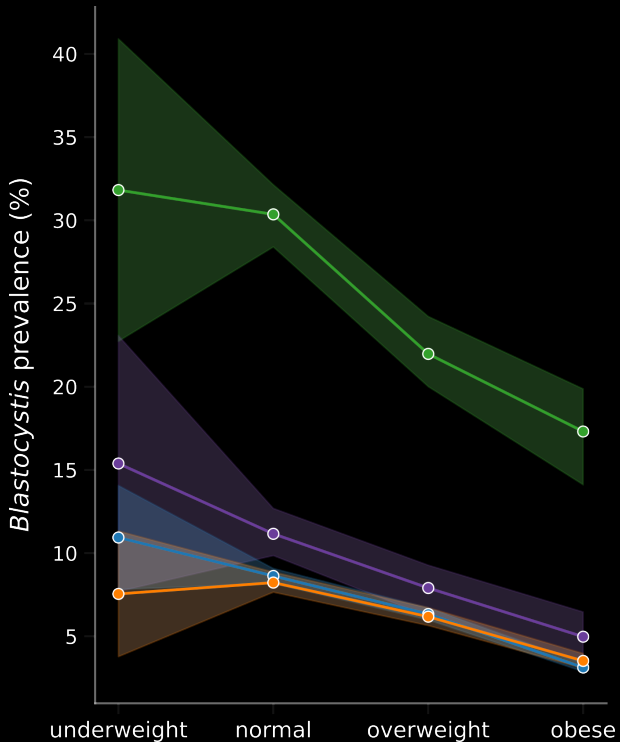
# BMI strongly linked in all PREDICT cohorts



# BMI strongly linked in all PREDICT cohorts

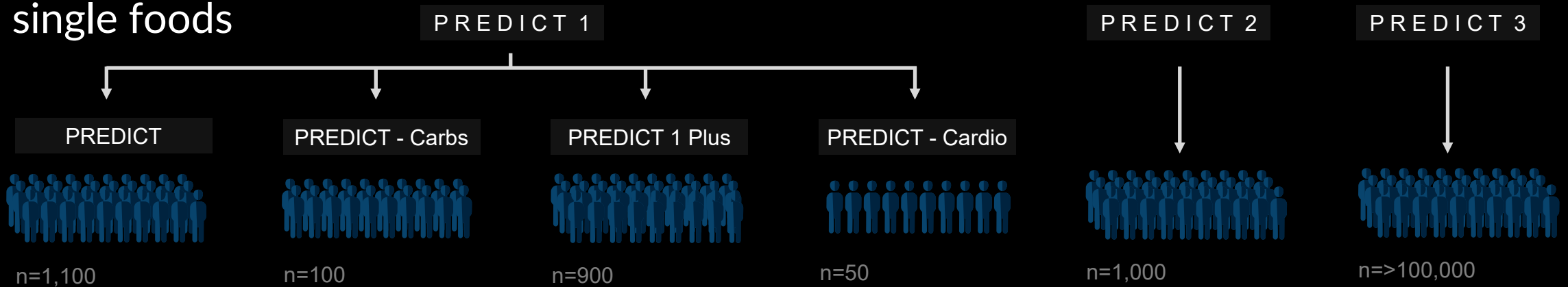
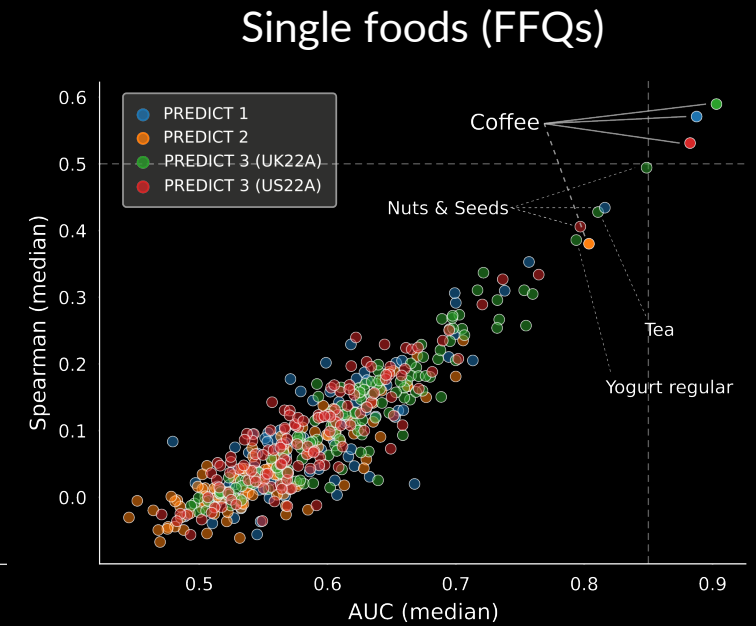
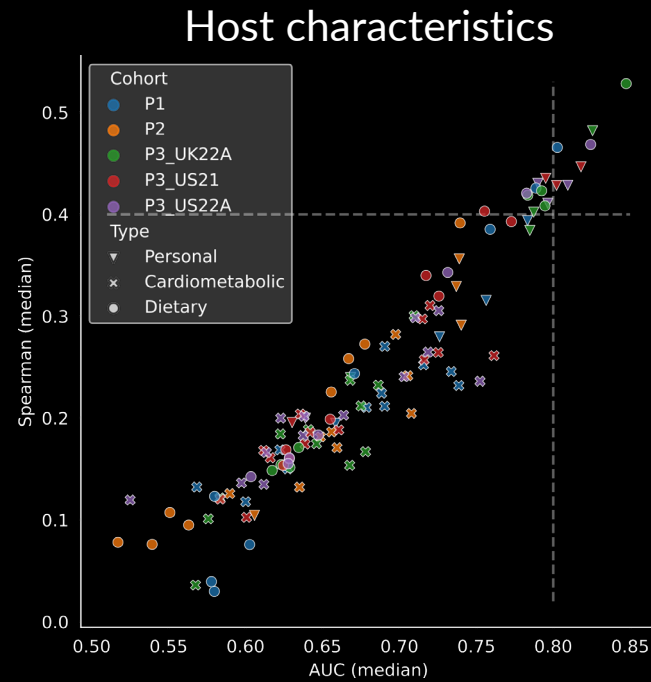


■ *Blastocystis* negative  
■ *Blastocystis* positive

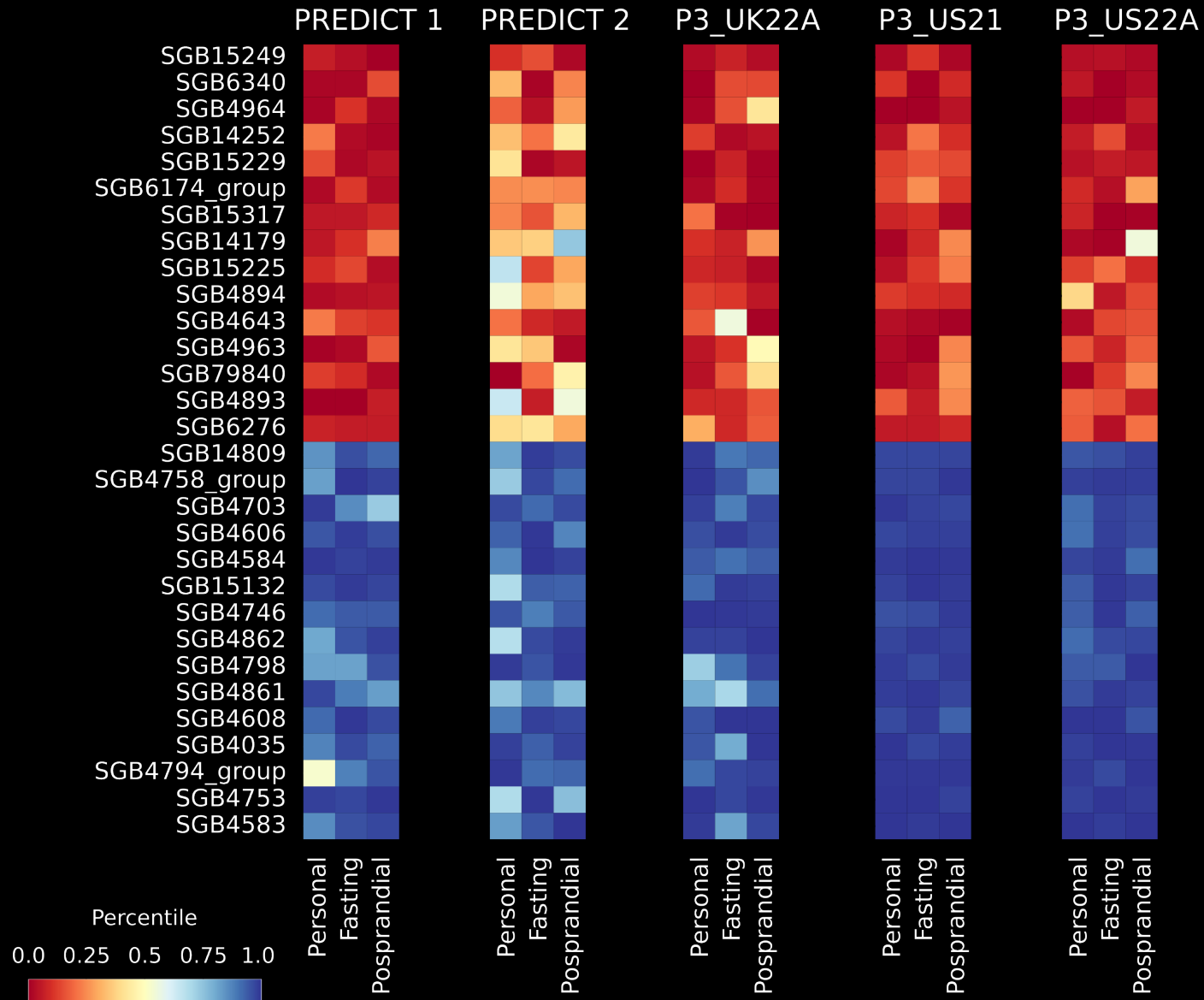


# Expanding to ~35k microbiome samples

- 5 PREDICT cohorts with ~35,000 microbiome samples
- Account for geography (UK, US)
- Stratify based on dietary patterns
- ML shows strong associations with host characteristics and single foods



# Rank SGBs according to their metadata associations



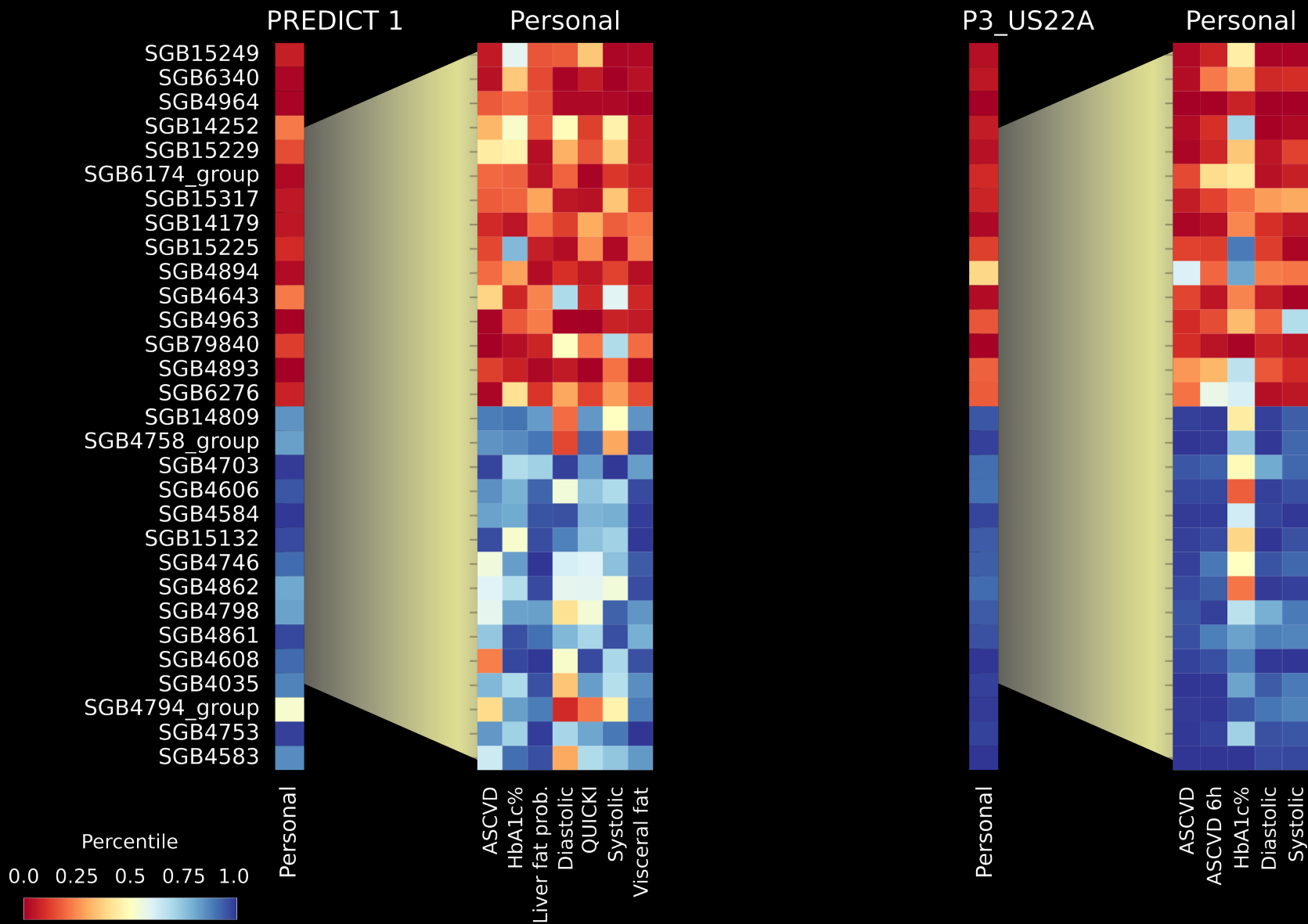
Metadata categories:

- Personal
- Fasting
- Postprandial

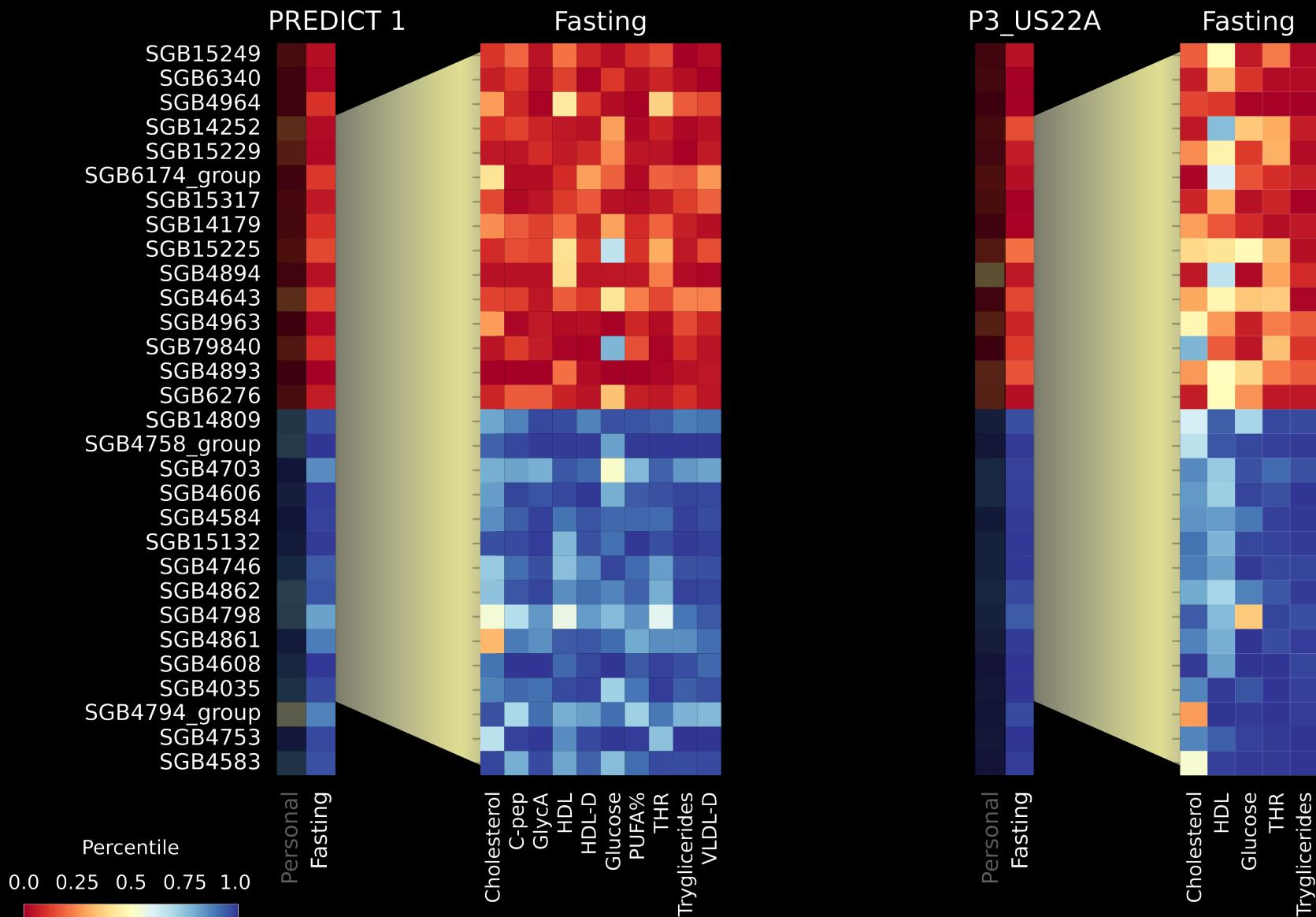
Each category contains several different markers

Average percentiles of ranked partial correlations for the 15 top and 15 bottom SGBs

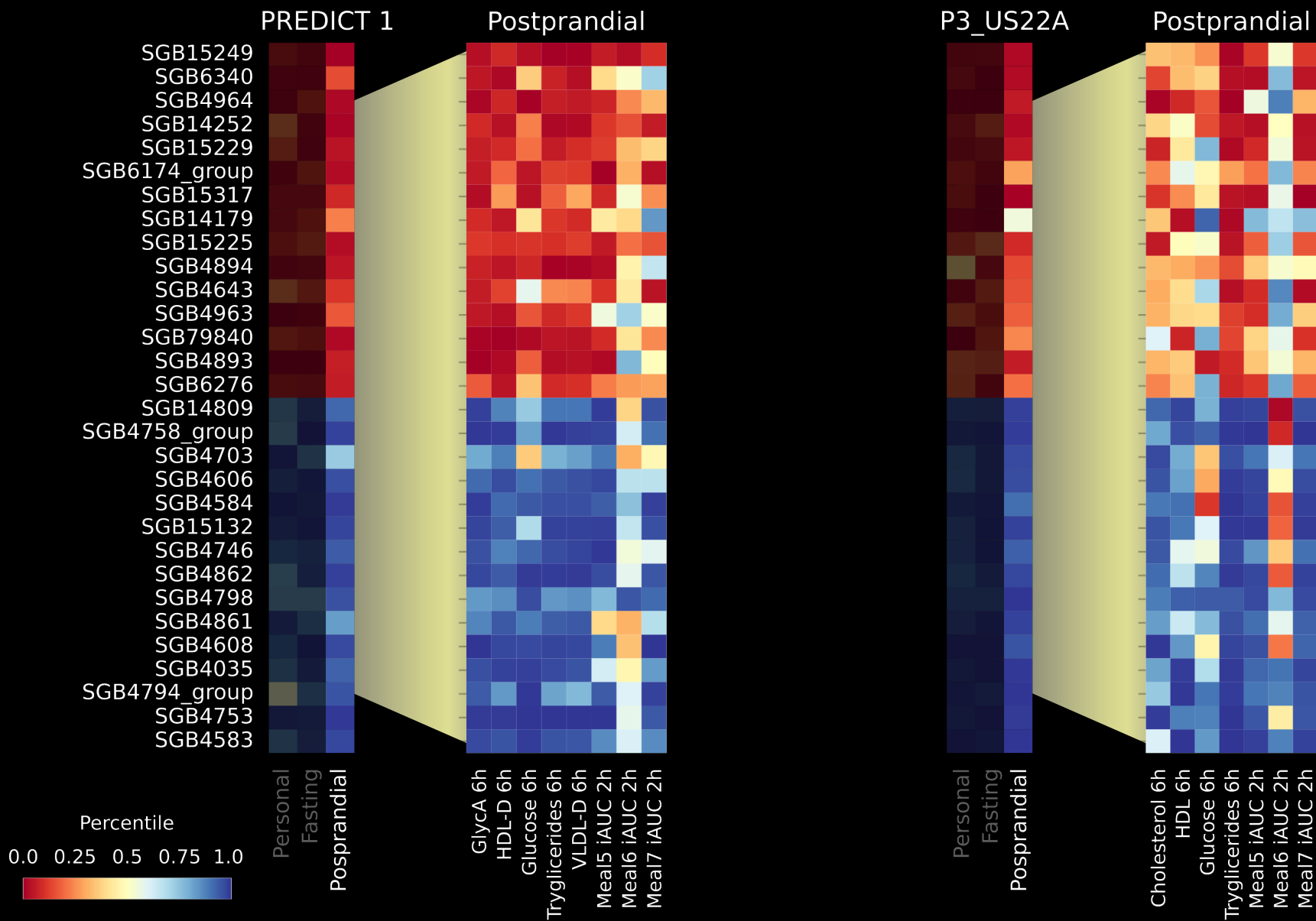
# Dive into the Personal category for UK and US



# Dive into the Fasting category for UK and US

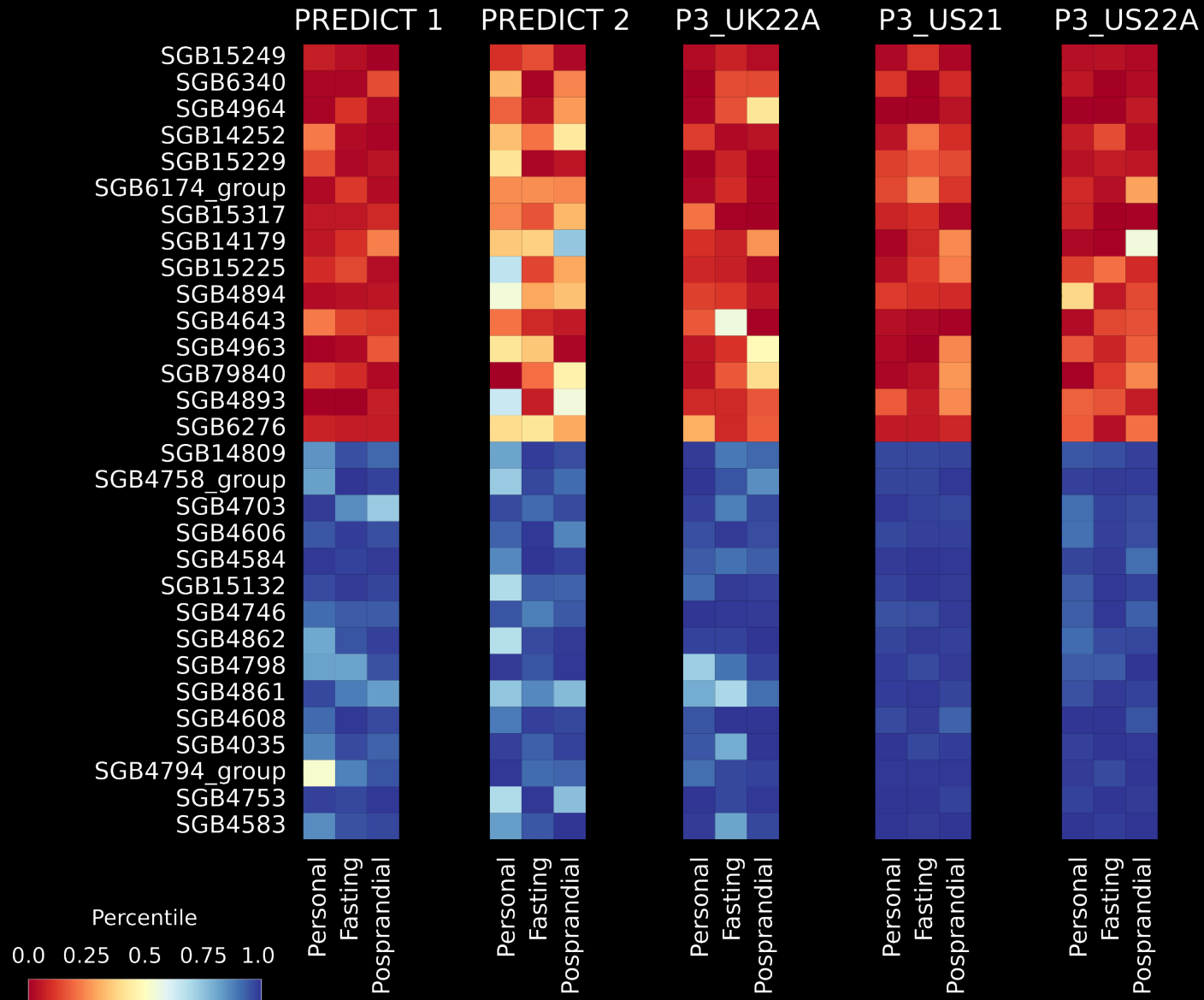


# Dive into the Postprandial category for UK and US

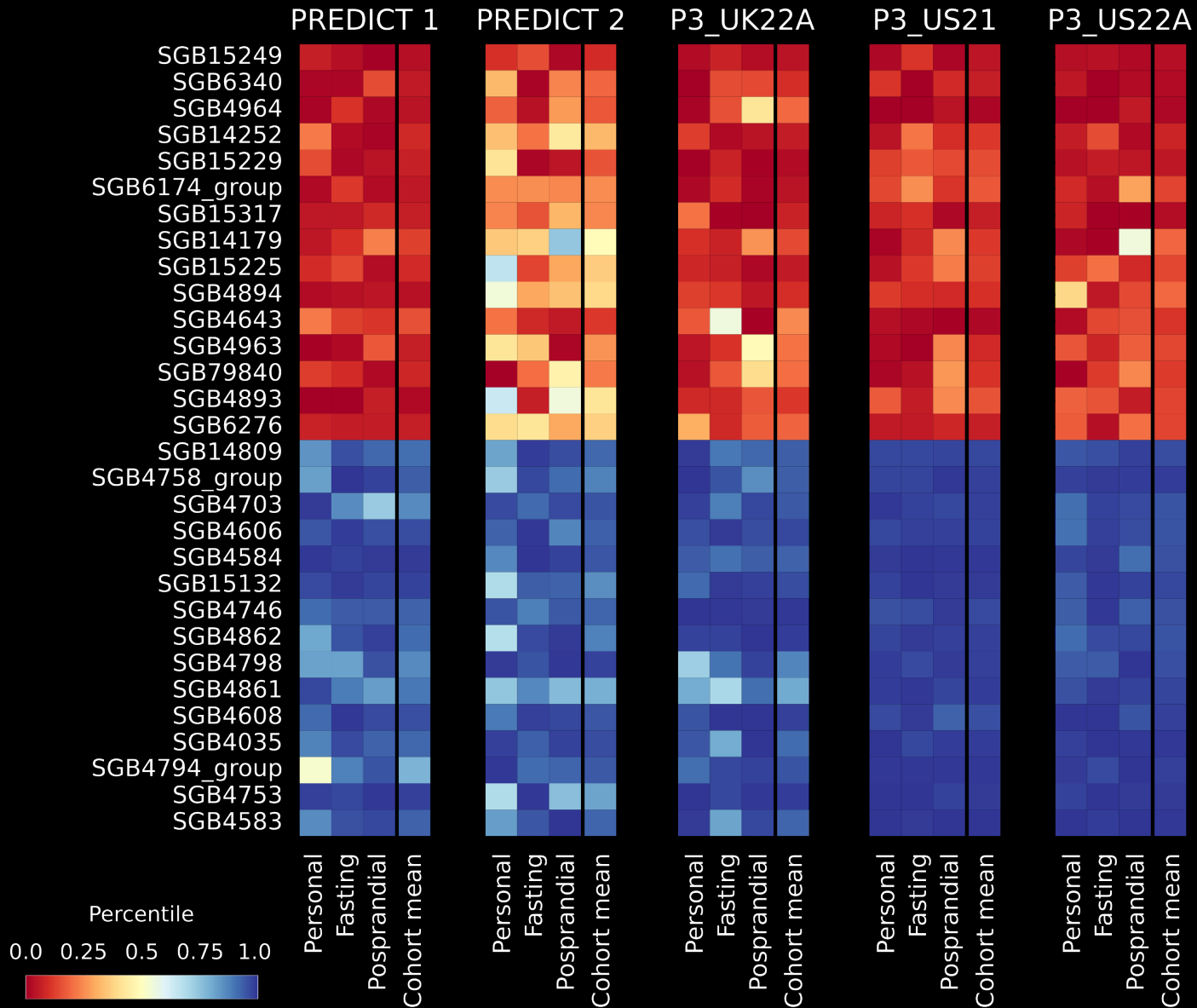




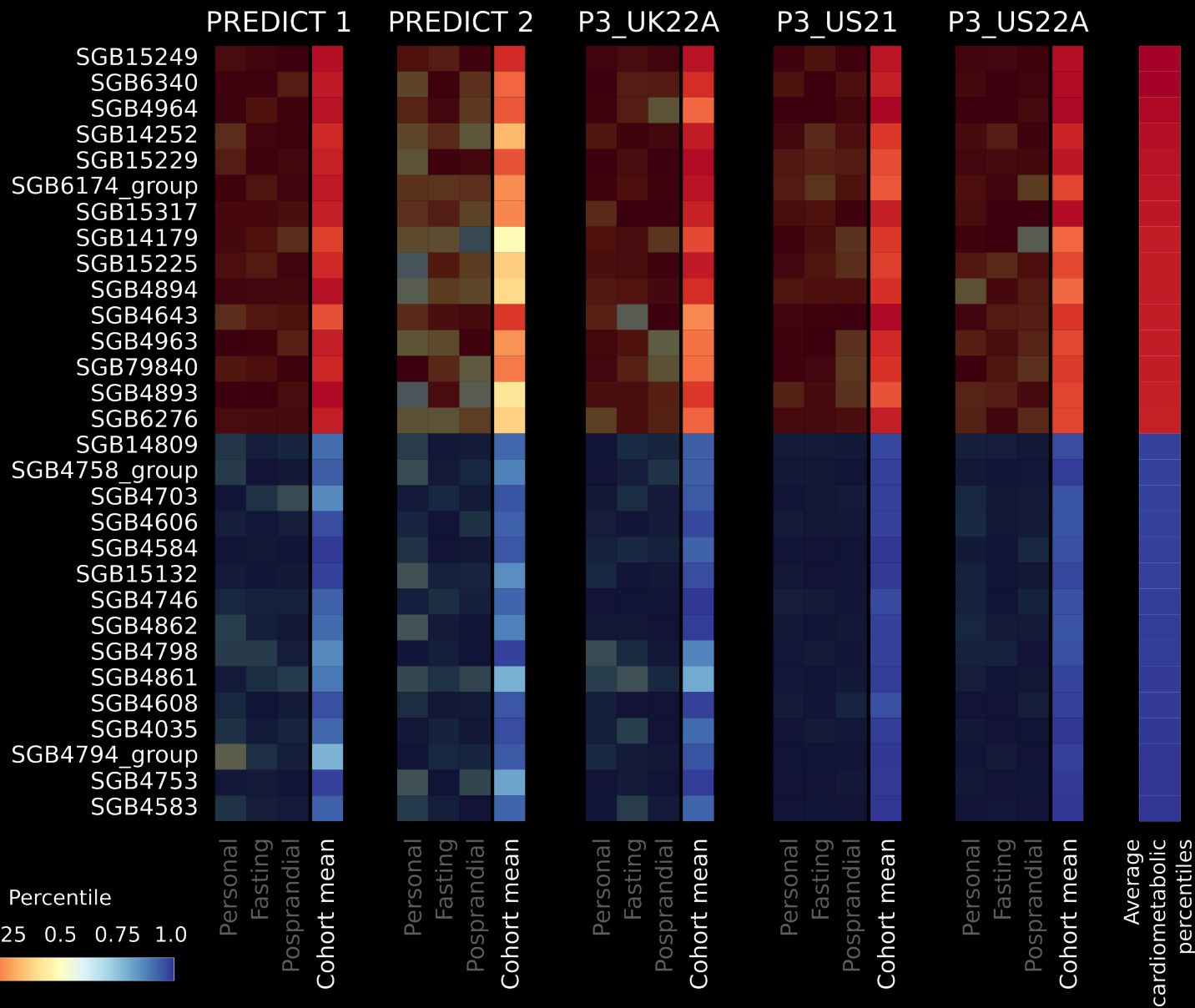
# Rank SGBs according to their metadata associations



# Cohort-level average SGB ranks

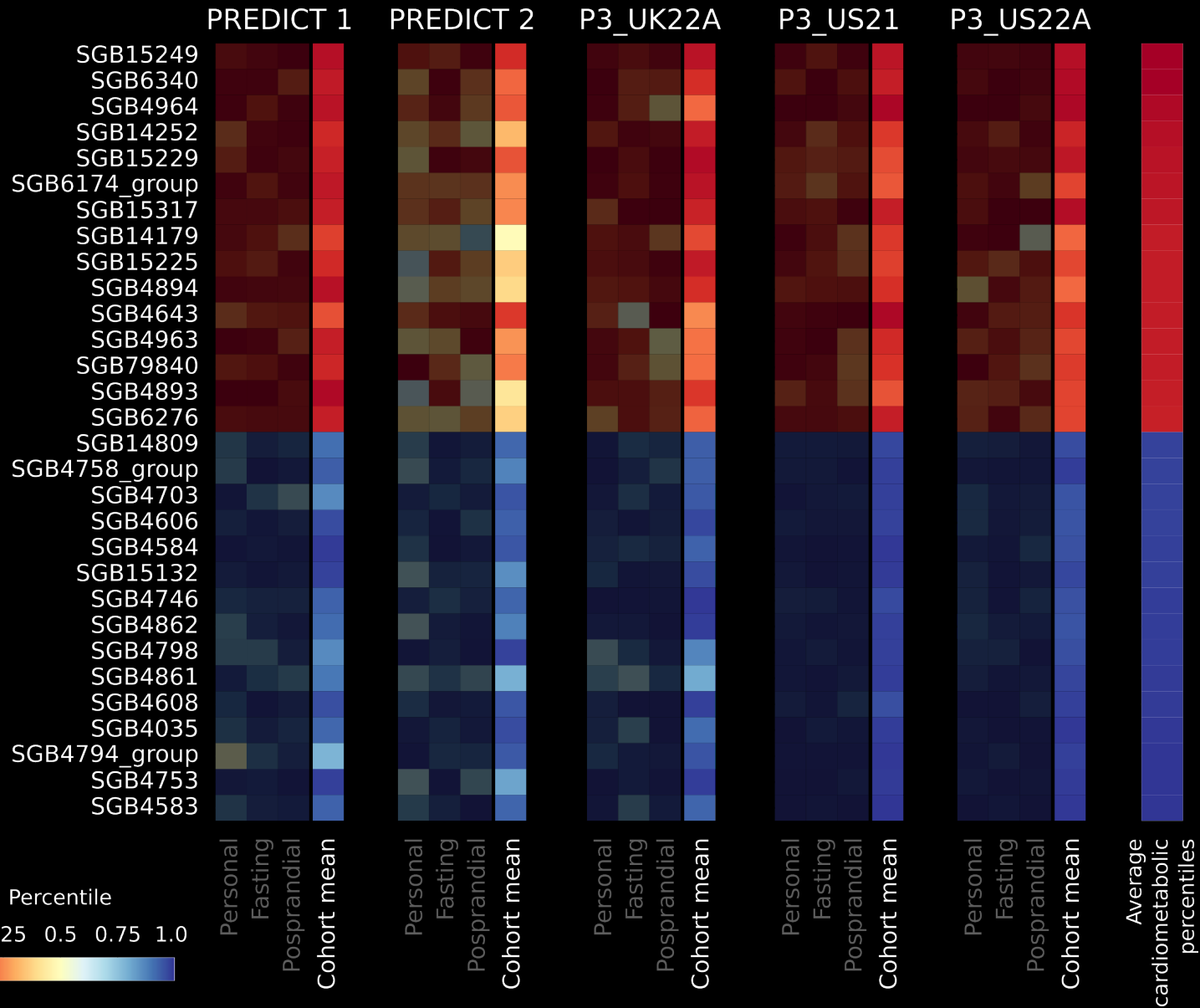


# Global cardiometabolic SGB ranks



We ranked in total 661 SGB species

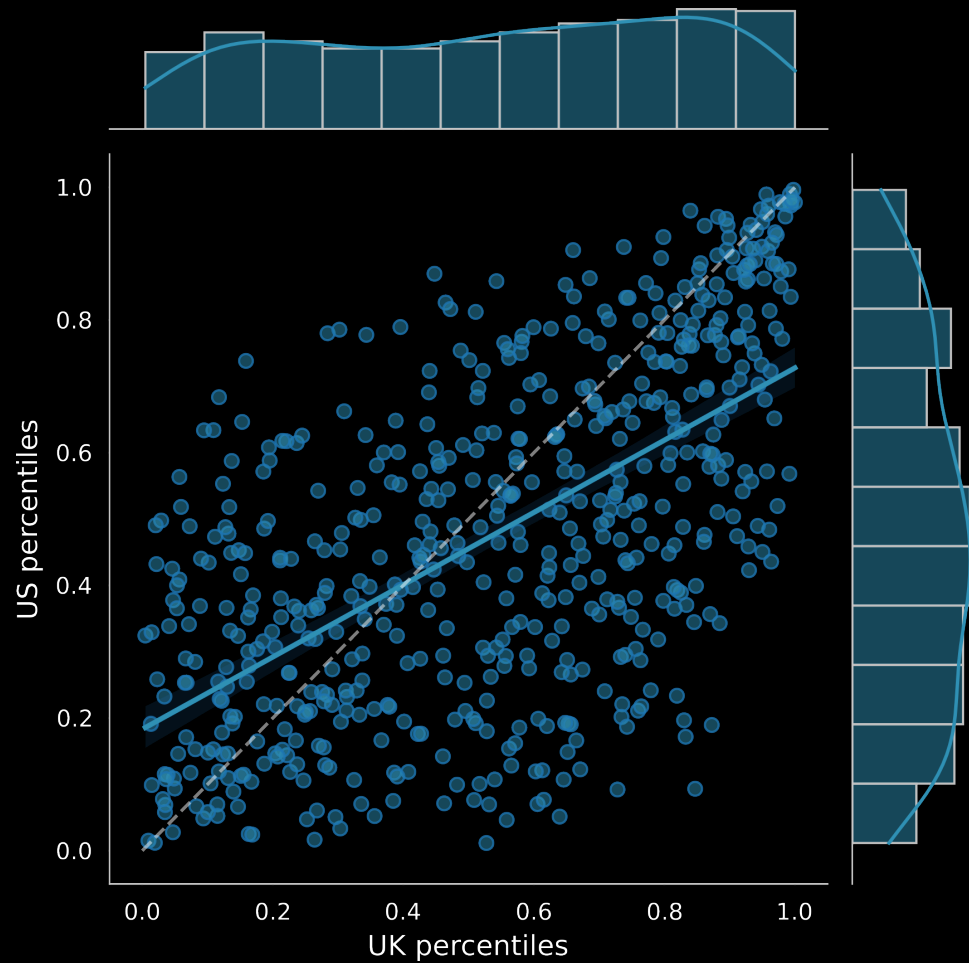
# Global cardiometabolic SGB ranks



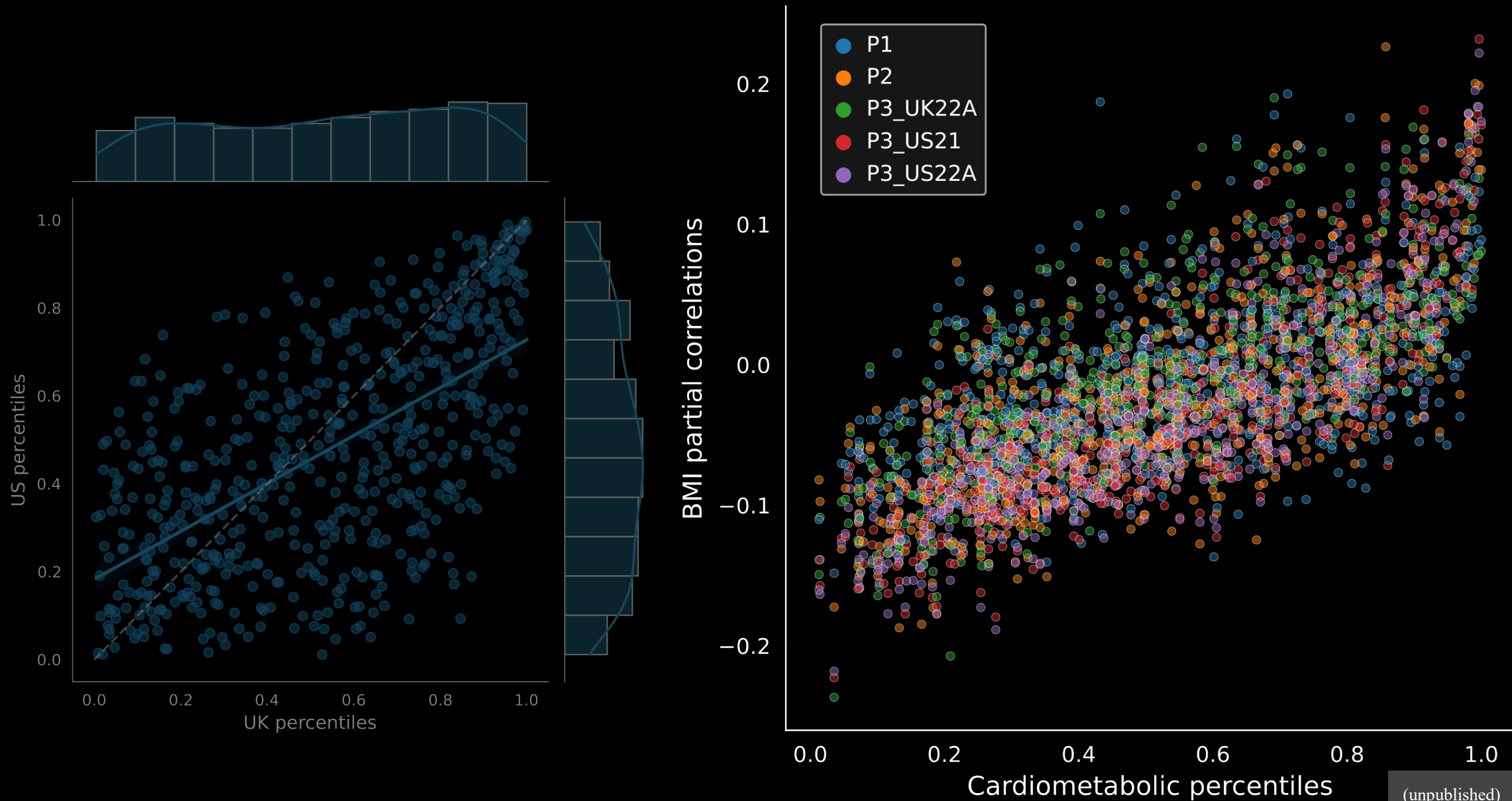
We ranked in total 661 SGB species

# Cardiometabolic ranks across geography

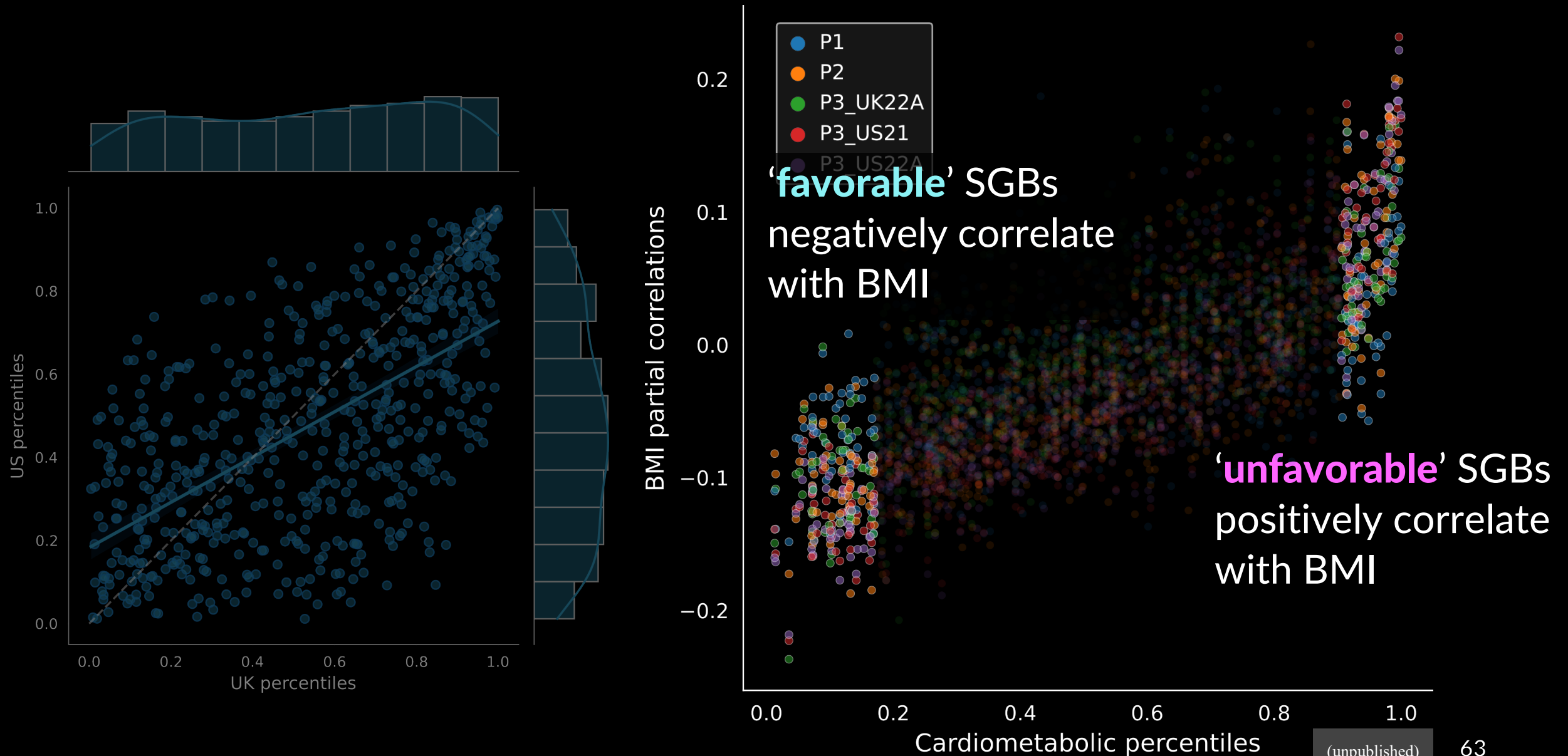
---



# Cardiometabolic ranks across geography and BMI



# Cardiometabolic ranks across geography and BMI







**We are recruiting!!**

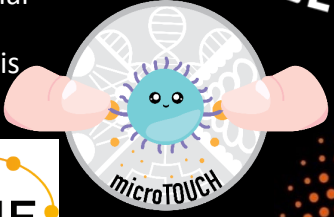
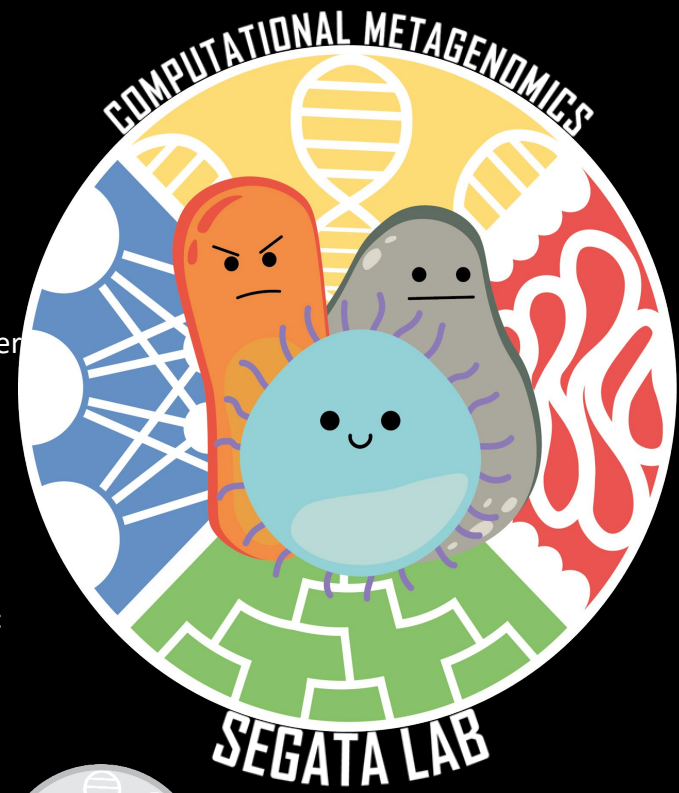
# Thanks!

 @cibiocm  
 @nsegata

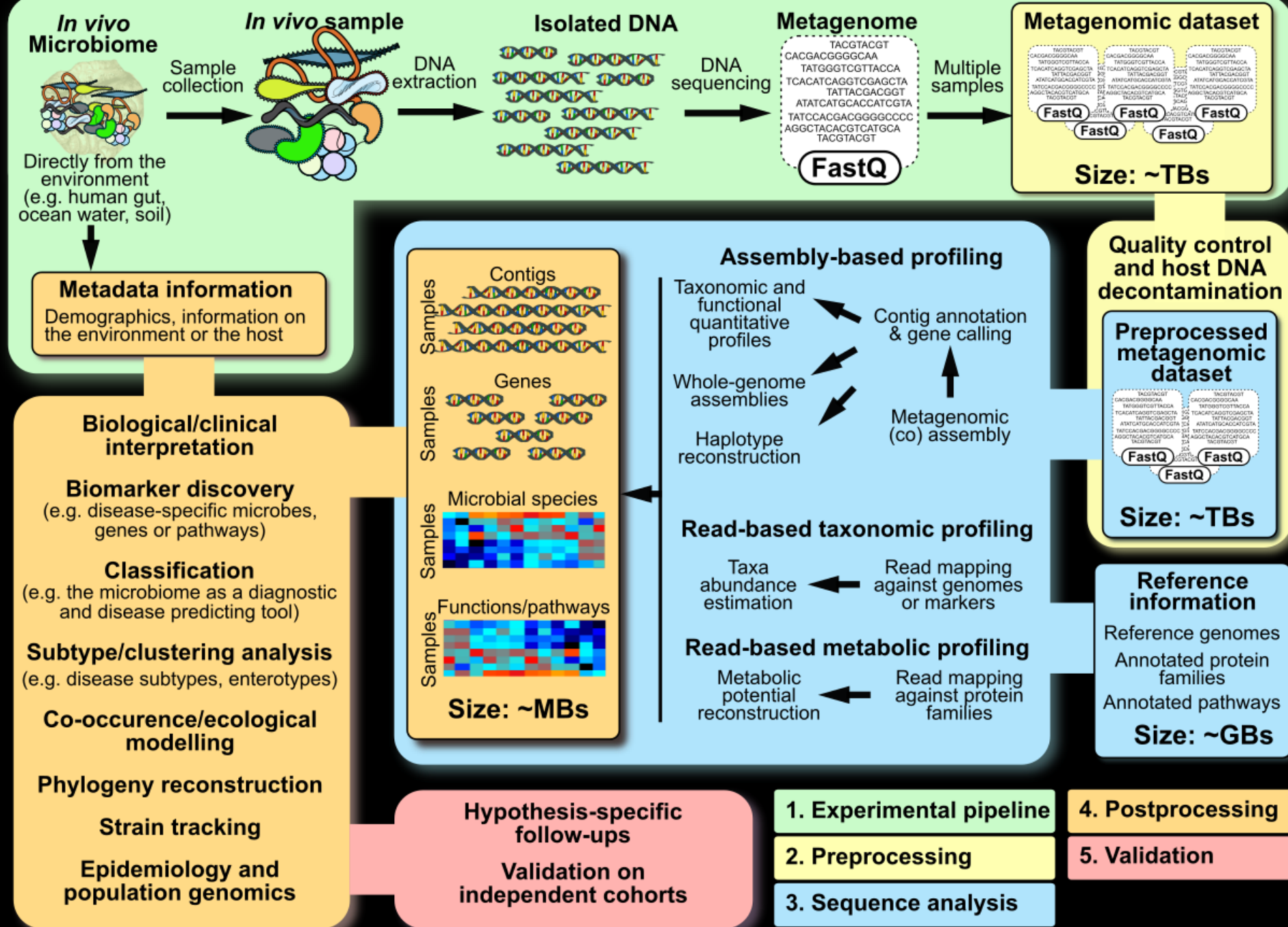
## The Laboratory of Computational Metagenomics



- Nicola Segata
- Francesco Asnicar
- Aitor Blanco-Miguez
- Gloria Fackelmann
- Serena Manara
- Paolo Manghi
- Liviana Ricci
- Mohamed S. Sarhan
- Marta Selma-Royo
- Sabrina Tamburini
- Mireia Valles-Colomer
- Federica Armanini
- Federica Pinto
- Giacomo Antonello
- Niccolò Carlino
- Matteo Ciciani
- Leonard Dubois
- Daive Golzato
- Katarina Mladenovic
- Amir Nabinejad
- Gianmarco Piccinno
- Elisa Piperni
- Michal Punčochář
- Andrea Silverj
- Charlotte Servais
- Paolo Ghensi



ACTIONS, MARIE CURIE, ISTITUTO G.B. MATTEI, EKLUND FOUNDATION, Horizon 2020, REPUBBLICA ITALIANA, M I U R, zoe, ONCOBIOME (Microbiota against cancer International research program), erc, erc, microTOUCH, MetaPG, SFAR (SIMONS FOUNDATION AUTISM RESEARCH INITIATIVE), FONDAZIONE CASSA DI RISPARMIO DI TRENTO E ROVERETO, LILT (LEGA ITALIANA PER LA LOTTA CONTRO I TUMORI prevenire e vivere), SEZIONE PROVINCIALE DI TRENTO, ITI (International Team for Implantology), SdP (Società Italiana di Parodontologia e Implantologia), TERME DI COMANO, LEO, LEONARDO DA VINCI



# The workflow of shotgun metagenomics

5-10Gb/sample

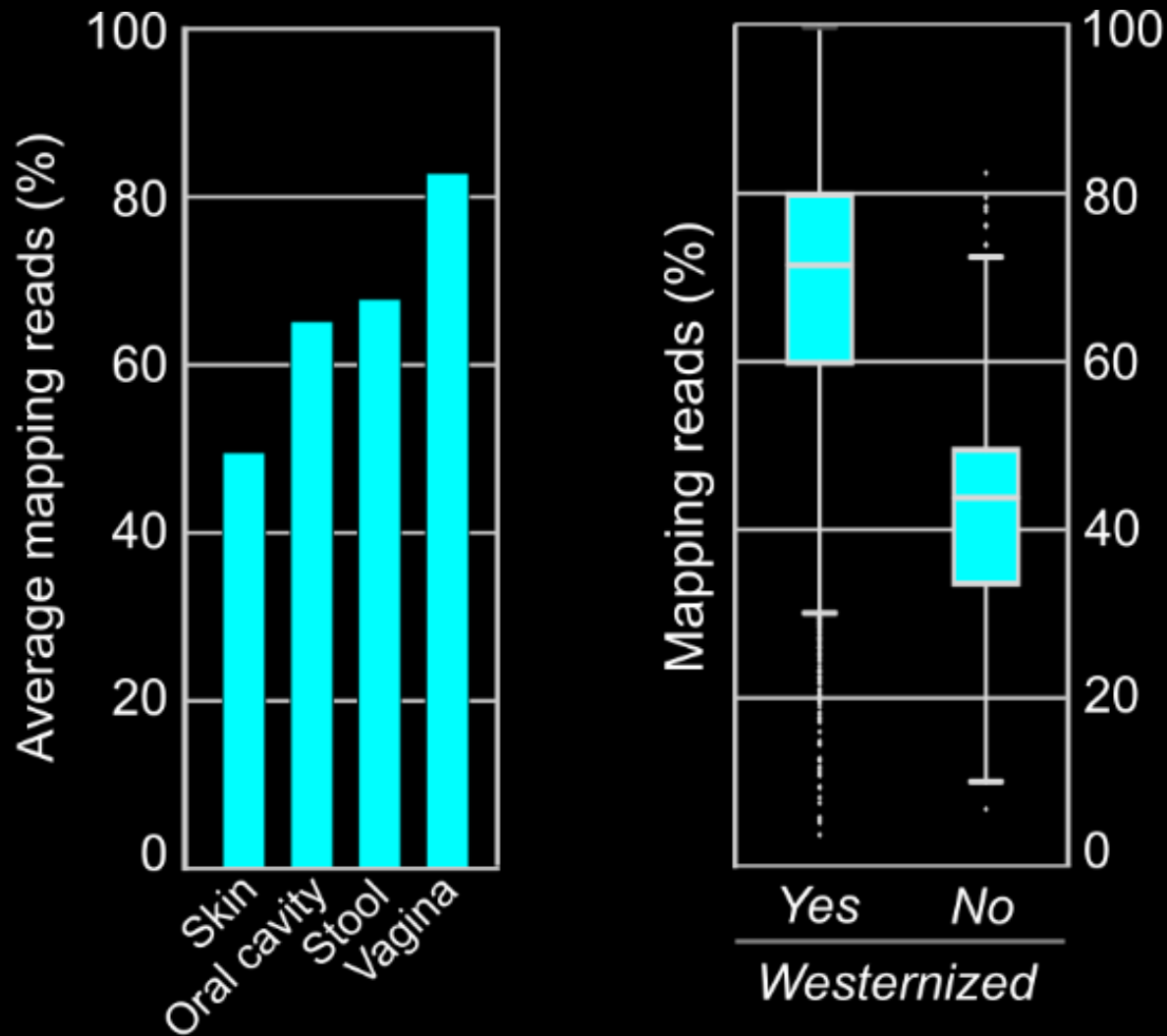
Our default setting:

- 384 samples on the NovaSeq S4
- 7.5 Gb/sample

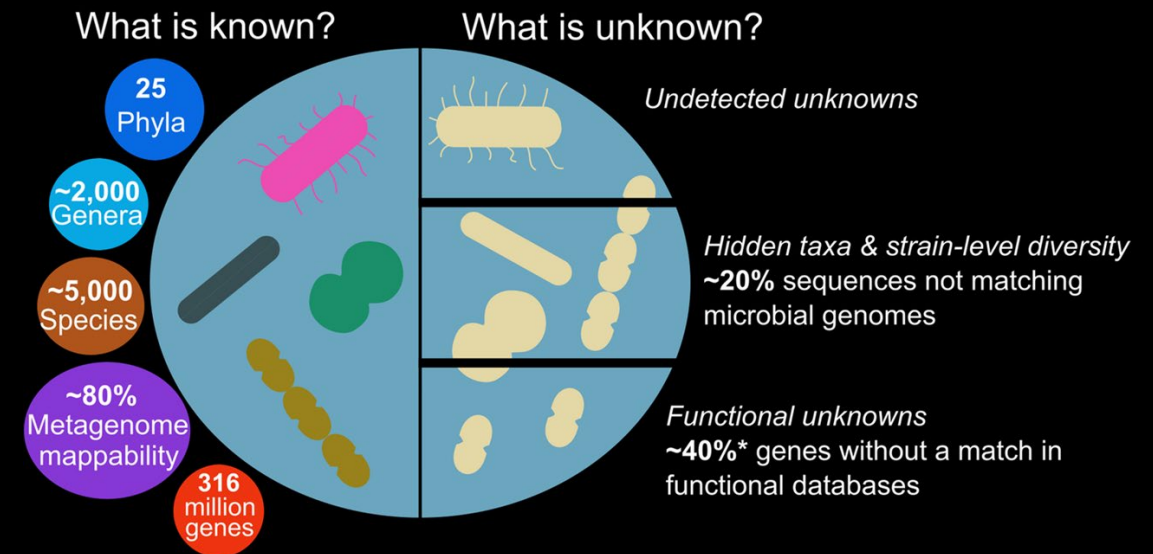
Human DNA:

- <3% in stool
- 70%-90% in saliva
- >95% in skin

# Many species are still uncharacterized



- Thousands of unknown species/strains?
- Millions of unsampled genes?
- Missing links with diseases/conditions?



Thomas & Segata, *BMC Biology*, 2019

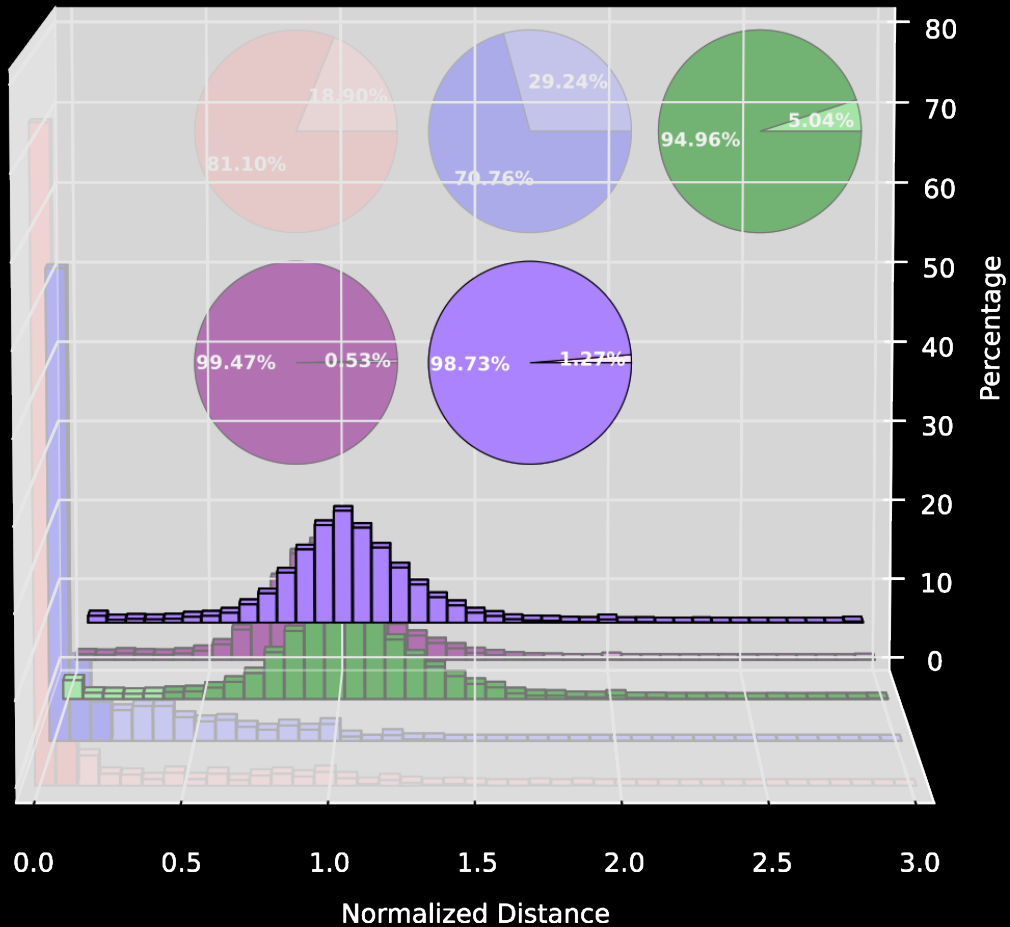
# Each human microbiome is unique at the strain level

- Subjects from around the world (~3000 sbj from 4 continents)
- Subjects from EU (6 countries)
- Subjects from US (from two universities)

Samples from same subjects collected at ~6 months

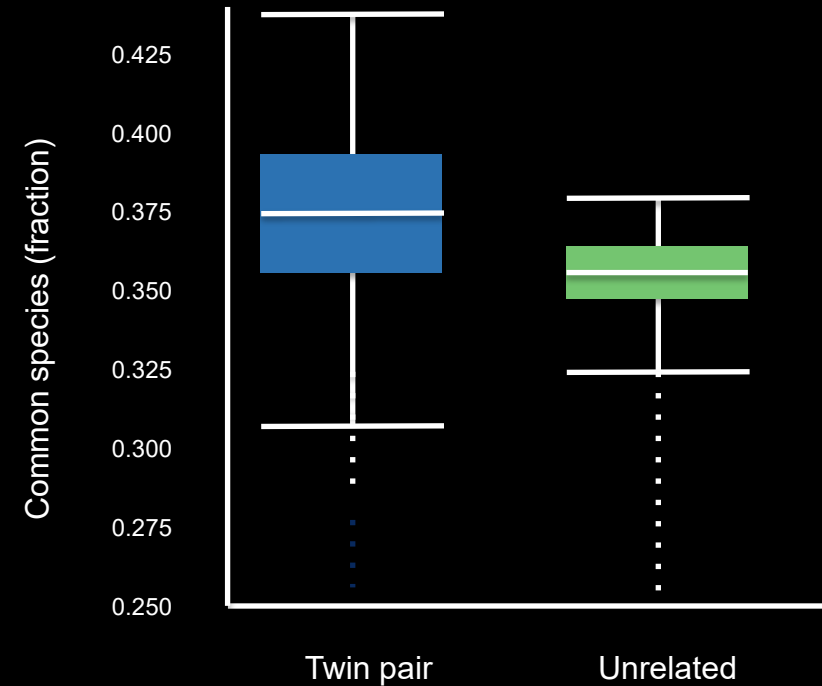
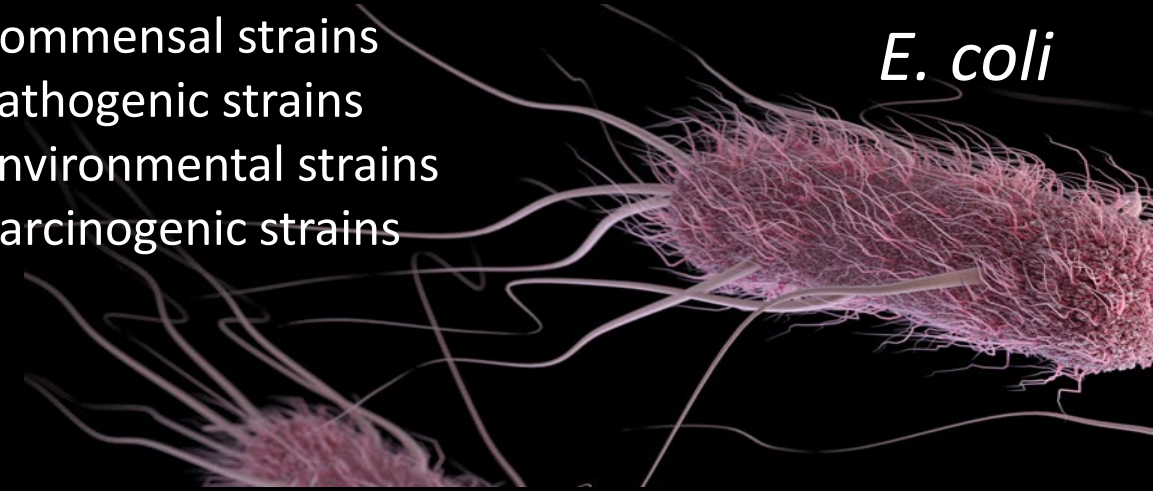
- Subjects from EU
- Subjects from US

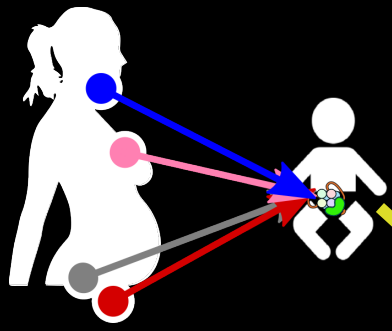
Truong *et al.*, *Genome Research*, 2017



Commensal strains  
Pathogenic strains  
Environmental strains  
Carcinogenic strains

*E. coli*

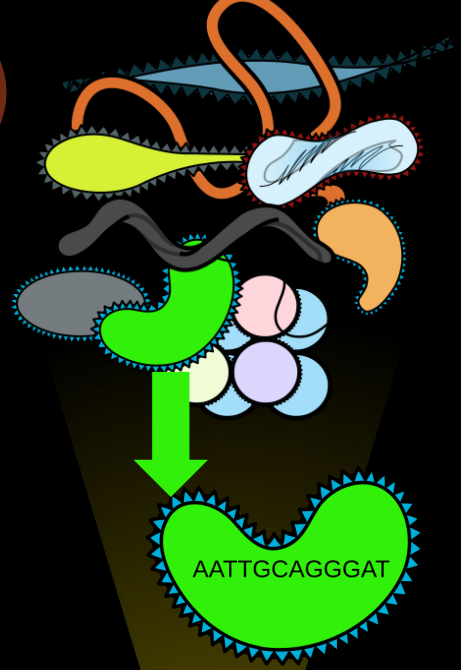




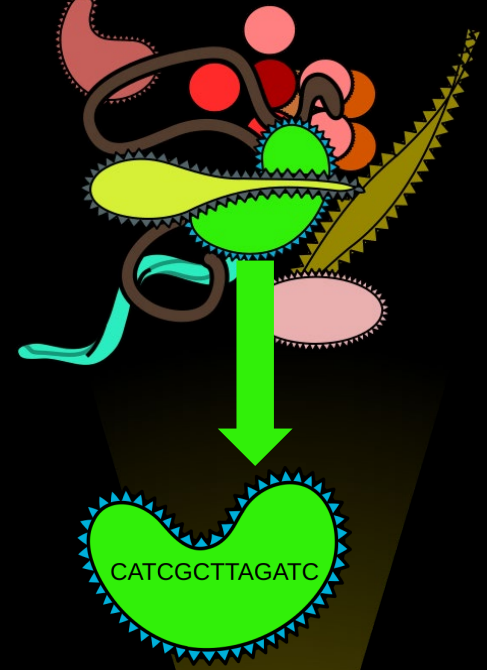
[Ferretti et al, Cell Host & Microbe, 2018  
Valles-Colomer et al, Nature, 2023]



4



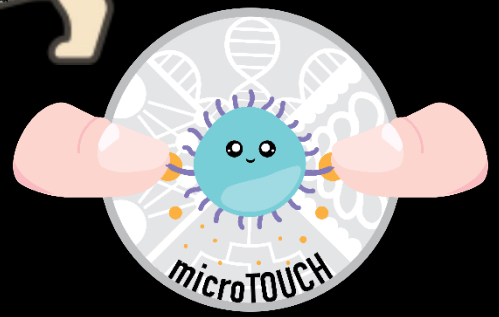
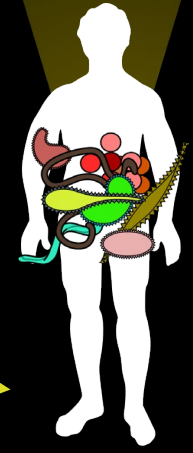
Average 35% shared species  
[Asnicar et al, Nature Medicine, 2021]



< 0.1% of shared strains  
[Truong et al, Genome Research, 2017]  
[Beghini et al, eLife, 2021]  
[Pasolli et al, Cell, 2019]



[Pasolli et al, Nature Communications, 2020  
Carlino et al, submitted]

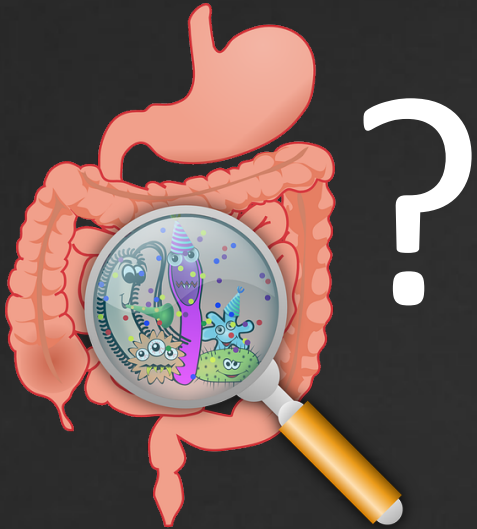


microTOUCH

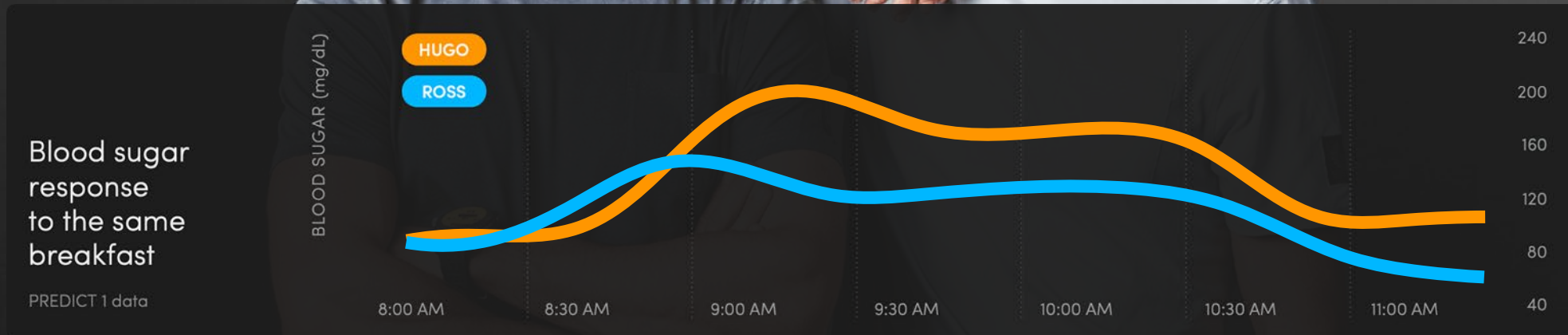
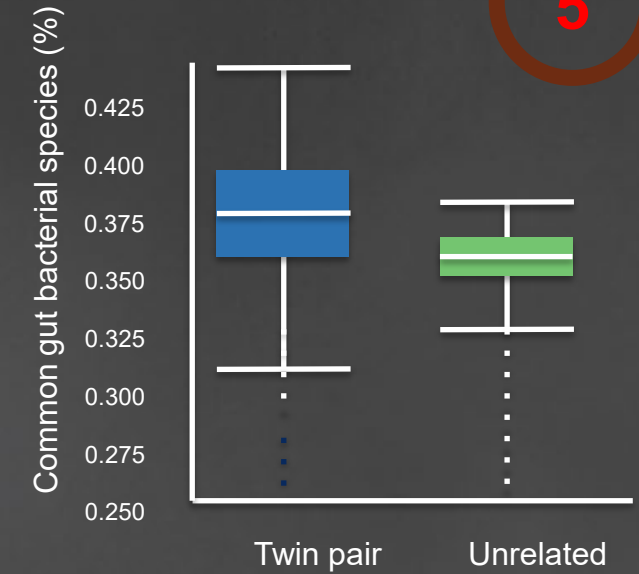
Hypothesis: extensive and intricate horizontal transmission

# We are all unique

...but the human microbiome is even more unique than us...  
...and could explain a lot of our phenotypic uniqueness!

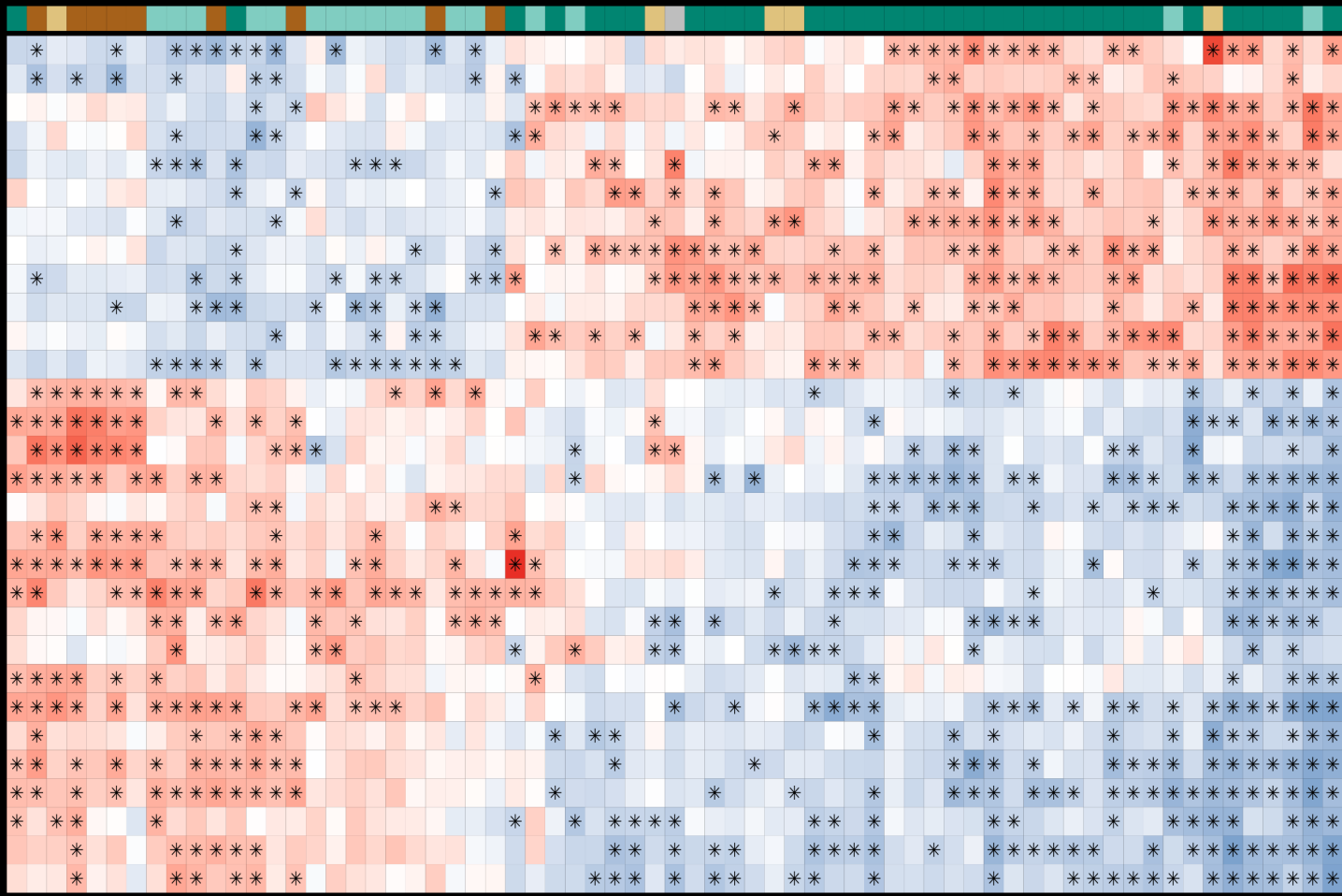


5

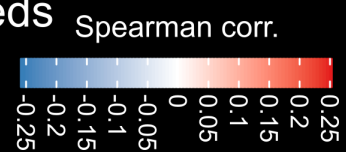


# PREDICT 1: a map of microbe-food associations

■ Healthy plant-based  
 ■ Less Healthy plant-based  
 ■ Healthy animal-based  
 ■ Unhealthy animal-based  
 ■ No category



Sauces    Beans    Fruit juice    Coffee    Eggs    Tomatoes    Fullfat yogurt    Seeds  
 Bacon    Savoury pies    Dairy dessert    Spinach



- Diets rich in **“healthy”, plant-based foods** associated with **“good”** gut microbes
- “Unhealthy”** microbe cluster was seen in those eating **“unhealthy” plant and “unhealthy” animal-based foods**
- Highlights the importance of food quality
- Overall dietary patterns matter

# Better characterization of the gut microbiome w.r.t. transit time: the blue poo



Asnicar *et al*, Gut 2021



## Questionnaire

Diet (FFQ)  
Stool form and frequency



## Microbiome analysis

Stool sample



## Blood samples

Fasting and post-prandial measures



## Transit time (hrs)

Blue muffins



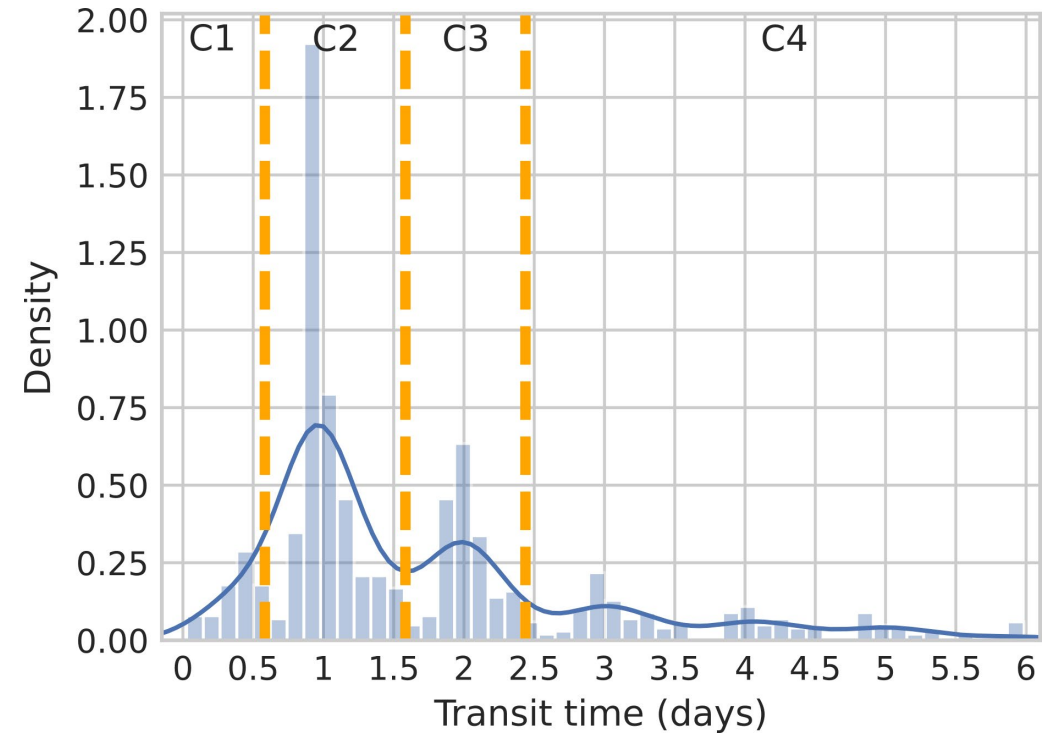
Blue Poo



Within 24hr prior to clinic visit

Clinic Visit

Self-report on mobile app



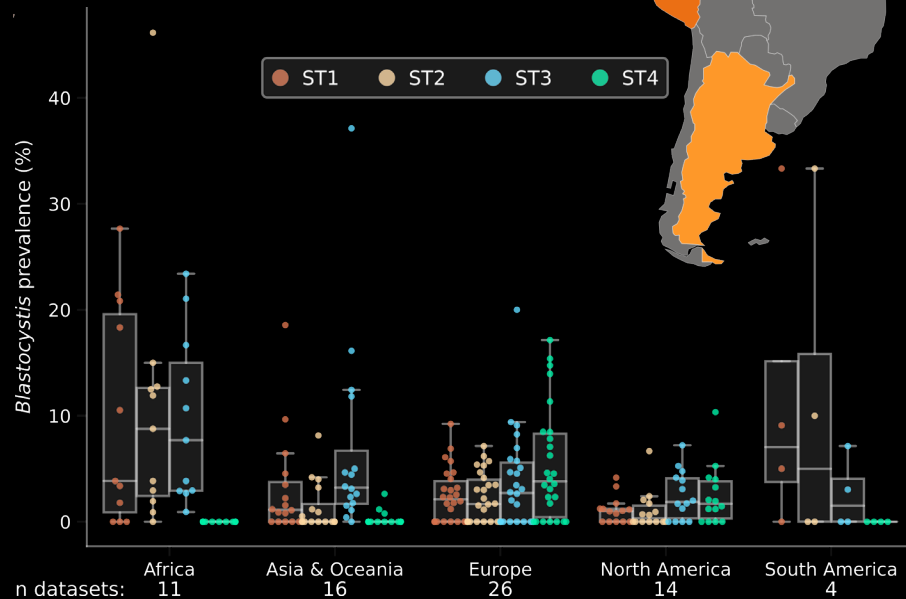
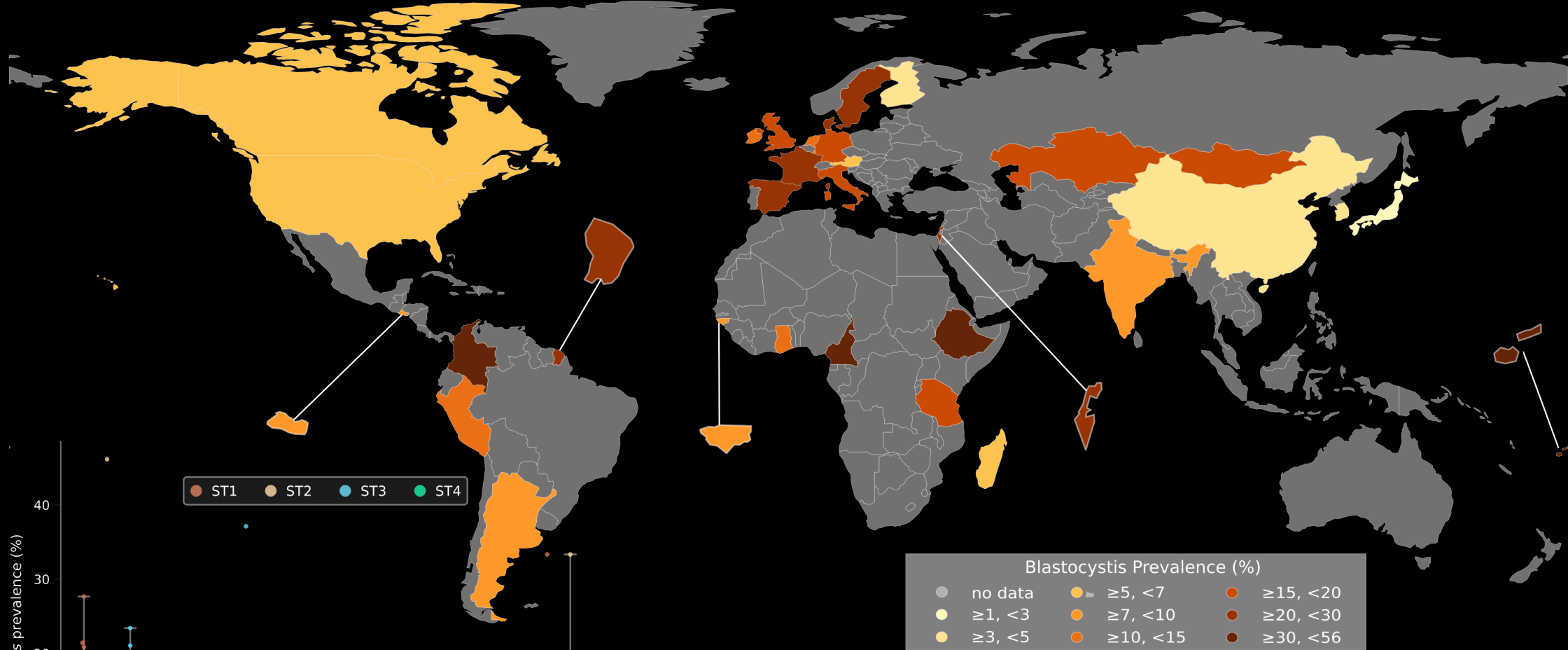


# Blastocystis is a prevalent “parasite”

8

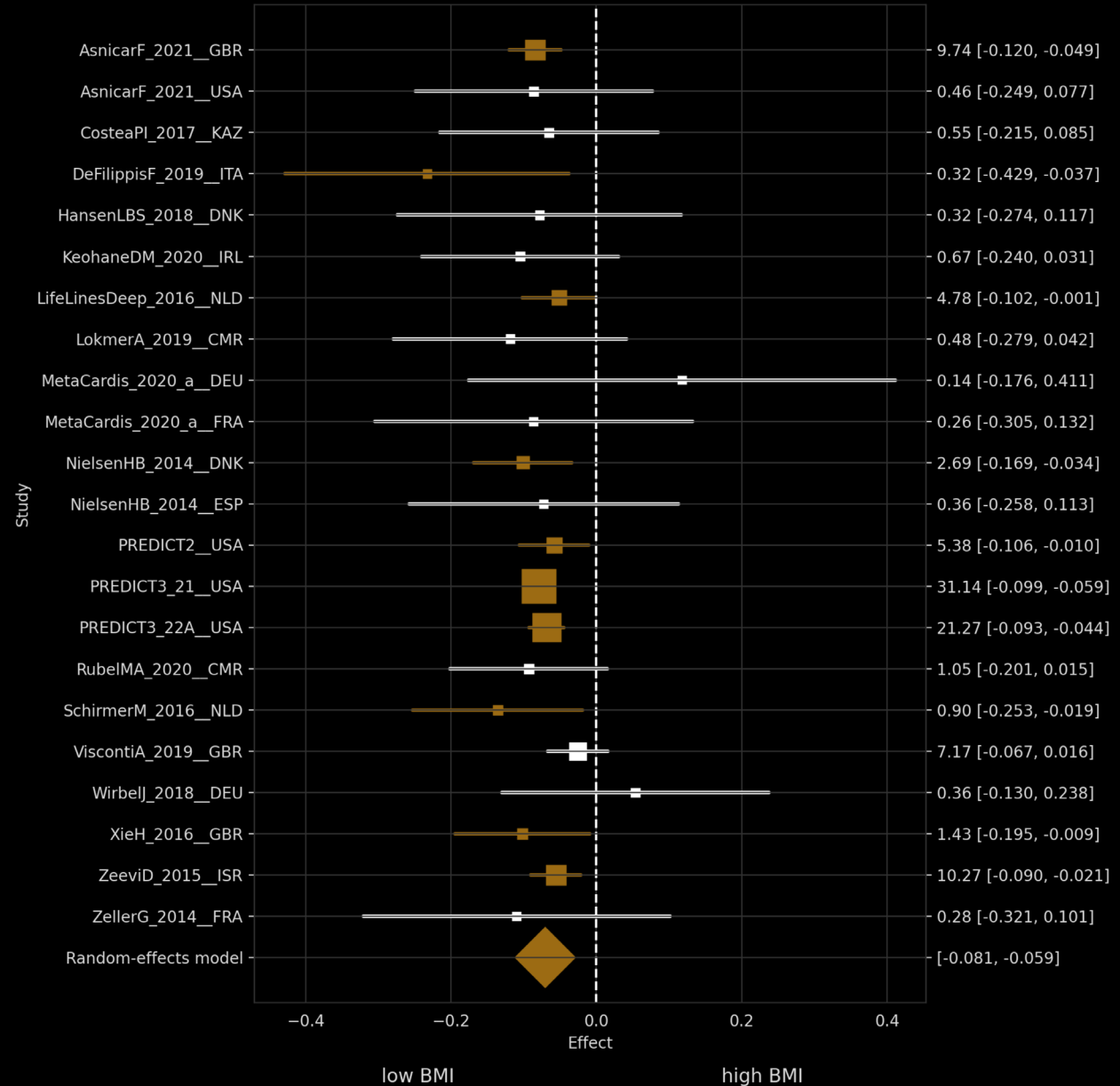
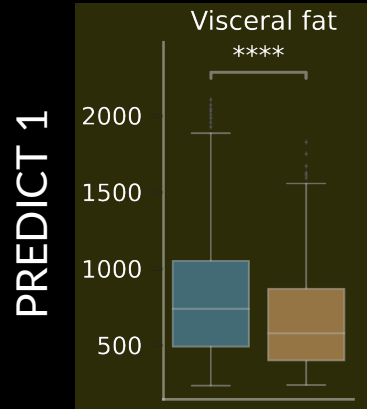


Elisa Piperni

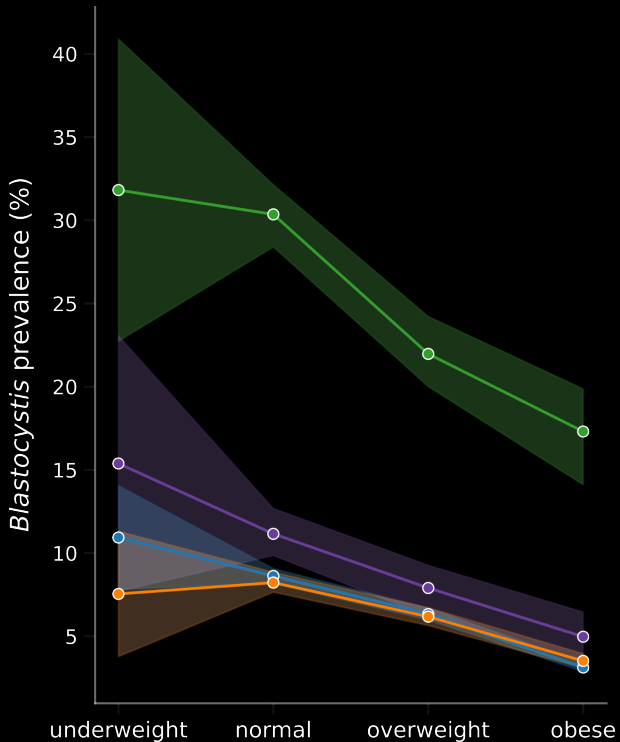


- Large differences in prevalence worldwide!
- ST3 specific to Westernized continents

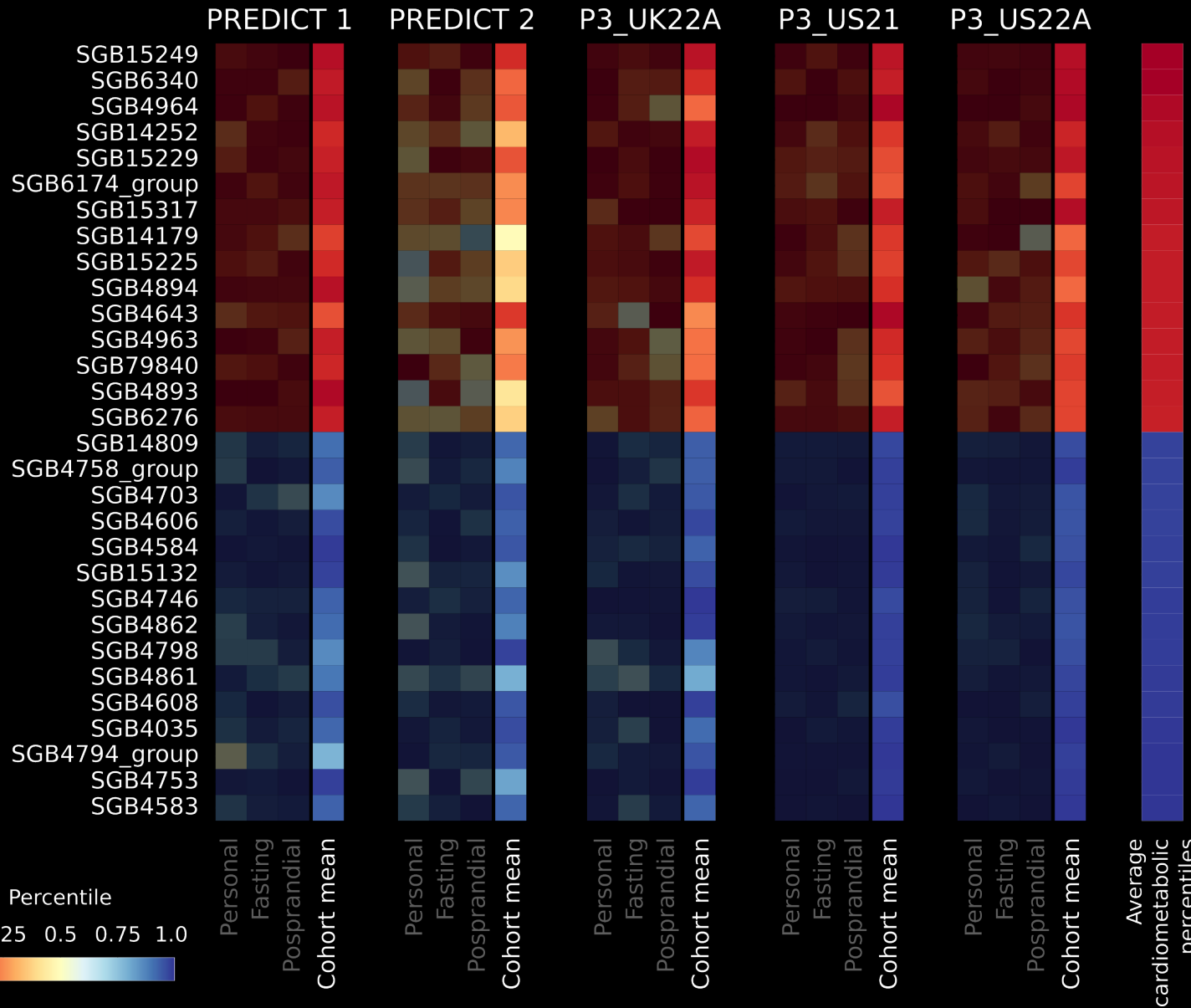
# BMI strongly linked in all PREDICT cohorts



■ *Blastocystis* negative  
■ *Blastocystis* positive



# Global cardiometabolic SGB ranks



We ranked in total 661 SGB species