

GLOBAL RESEARCH ALLIANCE

ON AGRICULTURAL GREENHOUSE GASES

LRG meeting

Dr Fernanda Godoy Santos



RUMEN Gateway Project (*RUmen Microbial gENomics) - Flagship project



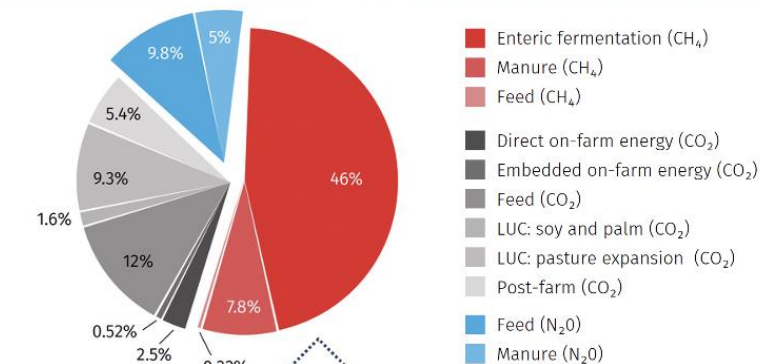
Culturing rumen microbes: a path to reduce methane

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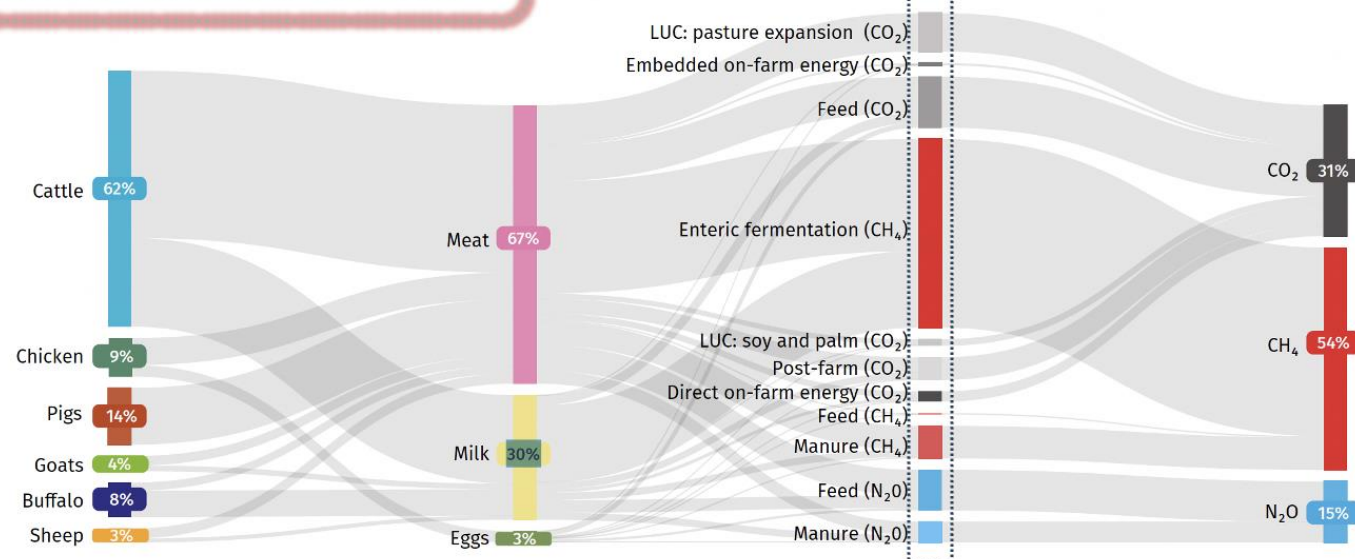
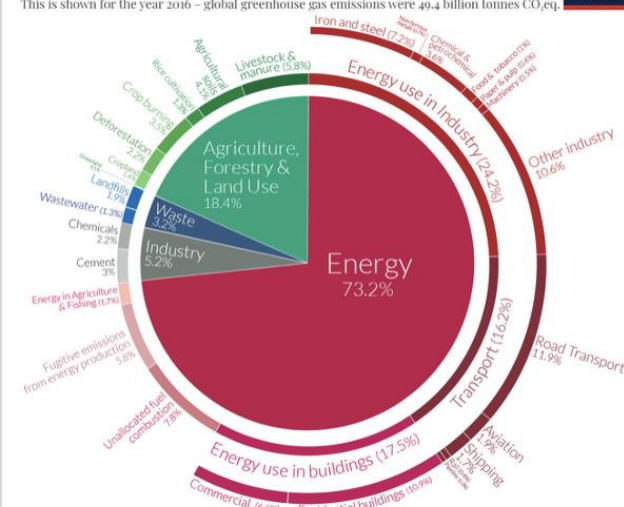
- Methane is a powerful heat-trapping gas.
- 50%-60% of methane emissions are the result of human activities.
- Ruminants contribute ~30% of anthropogenic methane.

Methane is produced in the rumen, an anoxic environment, as an end-product of microbial fermentation that serves to turn over excess H_2 in the rumen.



Global greenhouse gas emissions by sector
This is shown for the year 2016 – global greenhouse gas emissions were 49.4 billion tonnes CO₂eq.

Our World
in Data



FAO, 2023. Pathways towards lower emissions – A global assessment of the greenhouse gas emissions and mitigation options from livestock agrifood systems. Rome <https://doi.org/10.4060/cc9029en>

Culturing unlocks hidden microbial diversity

nature
biotechnology
OPEN

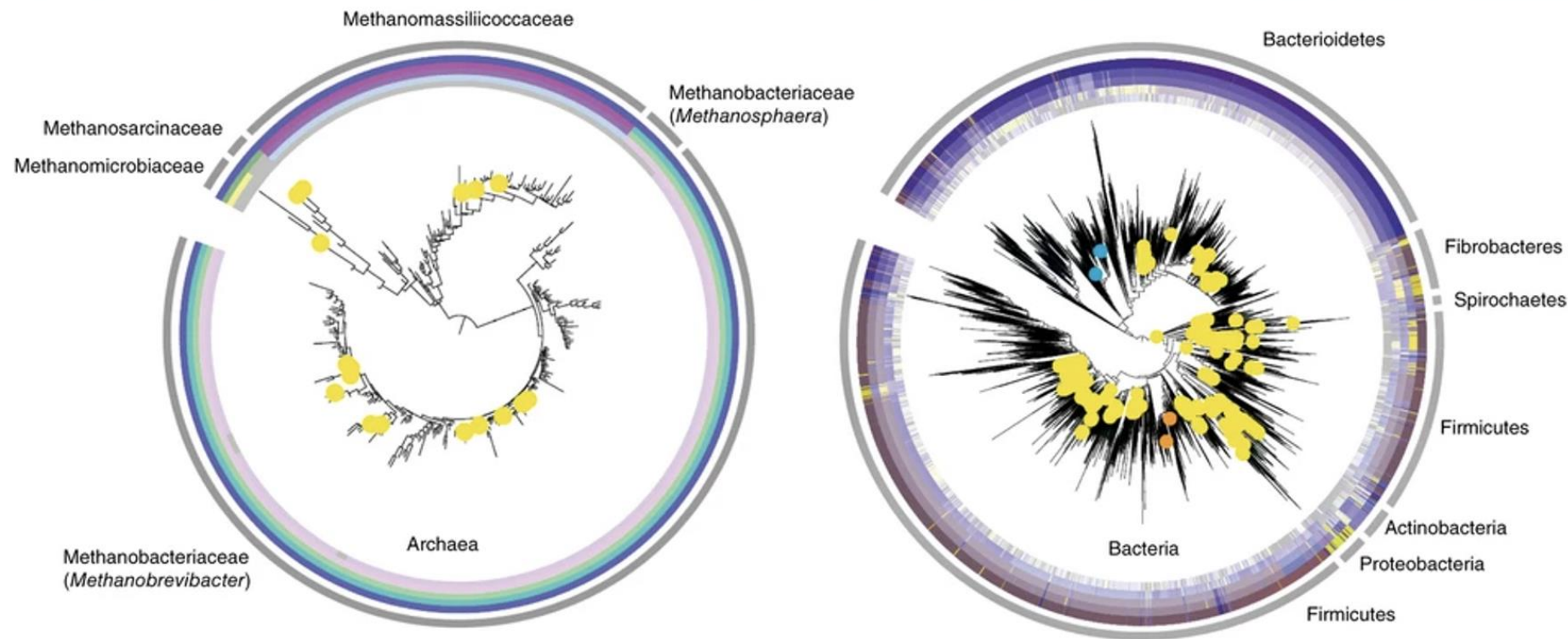
Cultivation and sequencing of rumen microbiome members from the Hungate1000 Collection

Rekhha Seshadri^{1,9,10}, Sinead C Leahy^{1,3,9,10}, Graeme T Attwood¹, Koon Hoong Teh^{1,8}, Suzanne C Lambie^{1,8}, Adrian L Cookson², Emily A Elor-Fadrush³, Georgios A Pavlopoulos¹, Michalis Hadjithomas¹, Neha J Varghese¹, David Paez-Espino¹, Hongate 1000 project collaborators¹, Rachele Perry¹, Gemma Henderson^{1,8}, Christopher J Creevey⁴, Nicolas Terrapon^{5,6,10}, Pascal Lapebie^{5,6}, Elodie Drula^{5,6}, Vincent Lombard^{5,6}, Edward Rubin^{1,8}, Nikos K. Kyrpides¹, Bernard Henrissat^{5,7}, Tanja Woyke¹, Natalia N Ivanova¹, William I Kelly^{1,8}

Productivity of ruminant livestock depends on the rumen microflora, which ferment indigestible plant polychaides into nutrients used for growth. Understanding the functions carried out by the rumen microflora is important for reducing greenhouse gas production by ruminants and for developing biofuels from lignocellulose. We present 410 cultured bacteria and archaea, together with their reference genomes, representing five cultivated rumen-associated archaeal and bacterial families. We evaluate polyphosphate degradation, short-chain fatty acid production and methanogenesis pathways, and assign specific taxa to these functions. A total of 136 organisms are functionally associated with the rumen. A detailed data set of rumen human gut microbiome data points, comparison with the human microbiome reveals specific enrichment for genes encoding de novo synthesis of pyruvate, B₁₂ encoding evolution by gene loss and potential vertical inheritance of the rumen microbiome based on underrepresentation of markers encoding essential genes. We estimate that our Hangege genome represents ~75% of the rumen bacterial and archaeal diversity in the rumen.

[illegible][illegible]

Received: 16 August 2017; accepted: 23 January 2018; published online: 19 March 2018; doi: 10.1039/c7cc06311a



Microbial community composition data from the Global Rumen Census overlaid with the 16S rRNA gene sequences (yellow dots) from the 501 Hungate catalogue genomes.

Project objectives

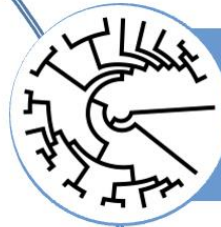
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Main goal: To uncover and characterise rumen microbial diversity through cultivation efforts, establishing a comprehensive and diverse microbial biobank to support innovation in sustainable livestock production.



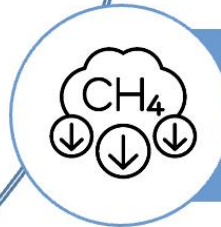
Supporting objectives:



Apply culturomics approaches across multiple microbial domains, including bacteria, archaea, and fungi.



Characterise pure and co-cultures phenotypically and genotypically to support functional and ecological analysis.



Develop a comprehensive, open-access database linked to the biobank to support innovation in methane mitigation.



RUMEN GATEWAY

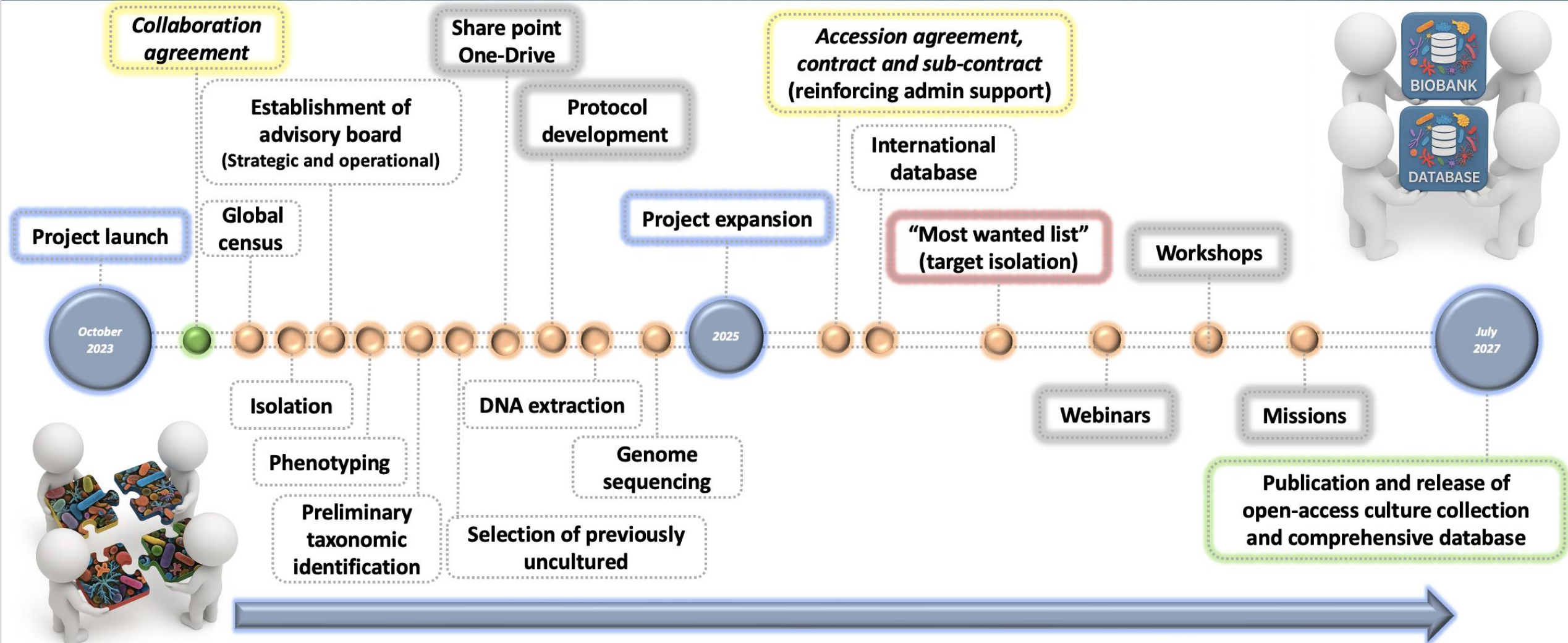
25 institutes (18 funded and 7 not directly funded by the project) | 39 PI | 10 Postdocs | 14 PhD students
6 continents | 17 countries | 27 time zones | 38 cities | 124 emails ... and counting!



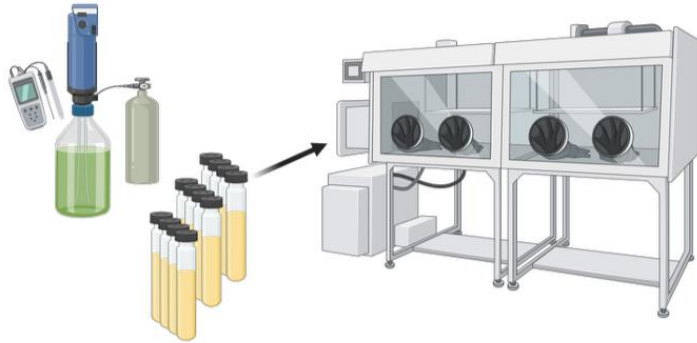
Project timeline

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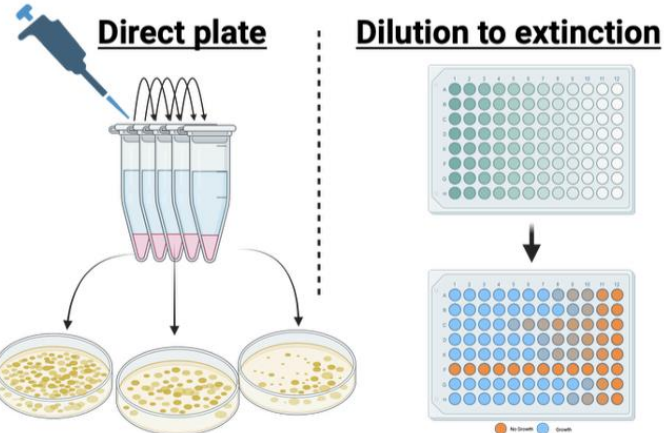
ON AGRICULTURAL GREENHOUSE GASES



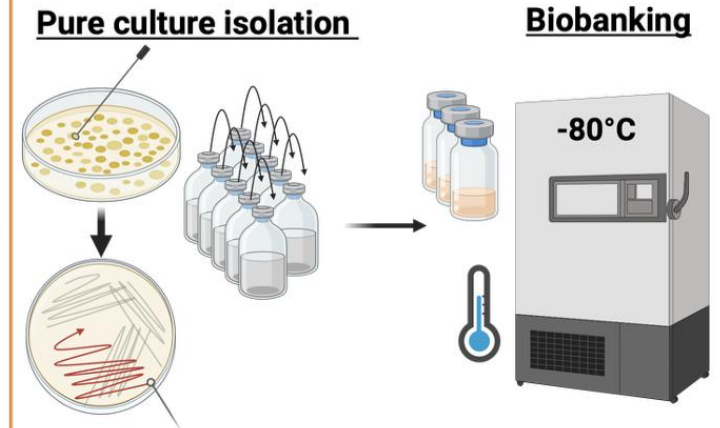
1. Sample collection and aliquoting



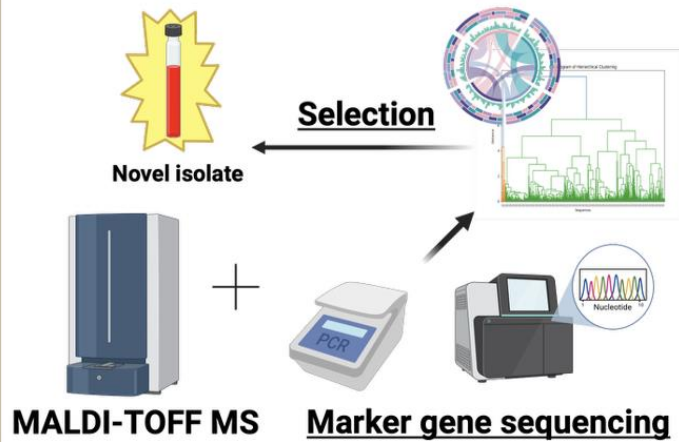
2. Distinct isolation



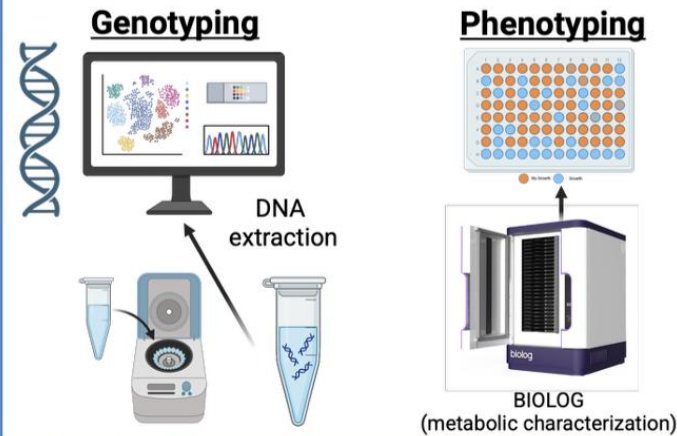
3. Purification and stocking



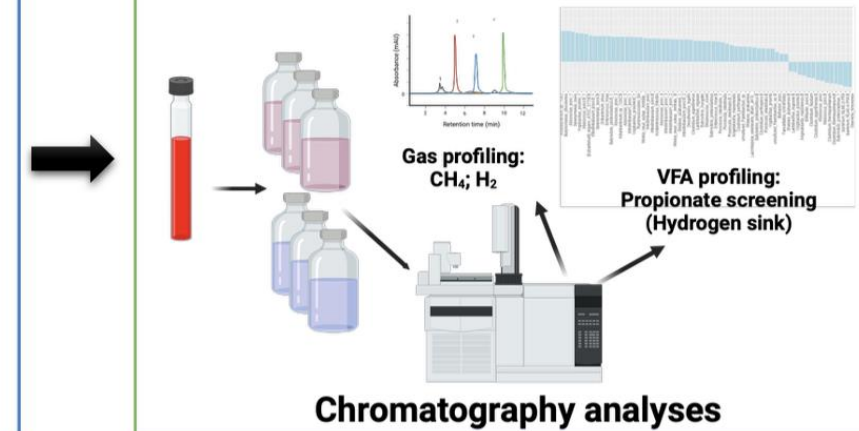
4. Preliminary taxonomic identification



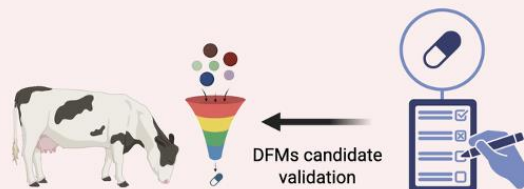
5. Isolate characterization

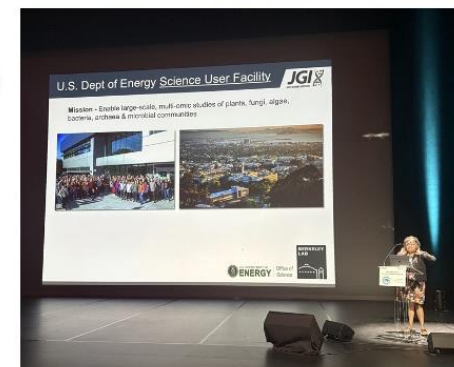
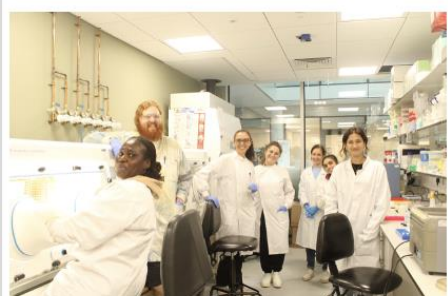
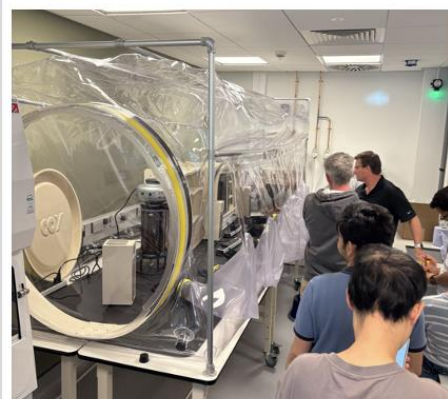
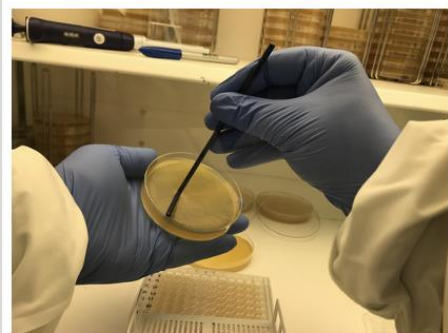


6. In vitro experiments



7. In vivo experiments





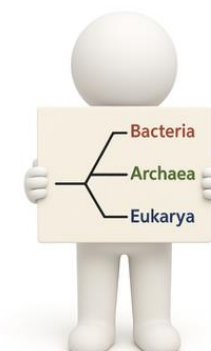
I UNIVERSITY OF
ILLINOIS
URBANA-CHAMPAIGN

 **QUEEN'S
UNIVERSITY
BELFAST**

INRAE



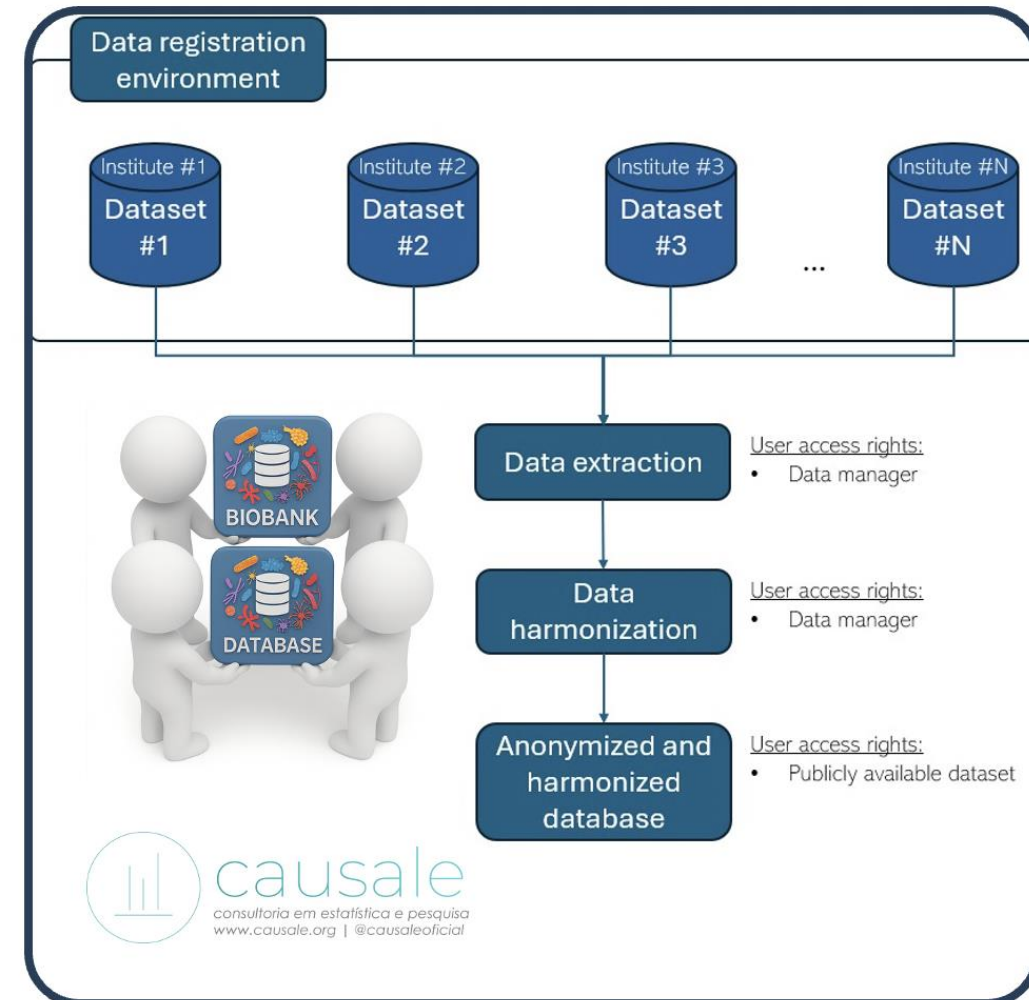
Hands-on training



JGI
JOINT GENOME INSTITUTE
DEPARTMENT OF ENERGY

Database management

- Create a **centralised database platform** that will serve as a comprehensive repository of metadata to support data integration across institutes.
- FAIR (Findable, Accessible, Interoperable, Reusable).
- Each institute will have its **own dataset** (its own silo), customizable according to the institute's needs.
- Relational links across institutes: samples, microbes, phenotypes, genomic resources.
- Supports structured data sharing, collaboration, and innovation in sustainable livestock technologies.



Culture collections*

Isolation medium

RM02, PC, M10; PYG, BHI, M2, Balch, L10, Orpin, AC B1, Columbia, Lowe's semi-defined, SS media.

**Including additional selective conditions and varied carbon and nitrogen sources.*

Isolation method

Direct plating, Dilution to extinction, Enrichment, Roll tubes, *In situ* isolation.

**Pure isolates and co-cultures kept in stock.*

Host

Bovine, Ovine, Caprine, Llama, Moose, Buffalo, Elk, Deer, Musk-ox, Bison, Camelid.

**Include varied breeds, ages, diets and native animals.*

Sequencing platforms

PacBio, Illumina, Nanopore, BGI-DNBSEQ, Sanger (marker genes).

***Numbers indicate total cultures held; availability may vary**



n = 673 (B); 8 (F)
gDNA = 87 (B); 8 (F)



AARHUS UNIVERSITY

n = 39 (B)
gDNA = 13 (B)



UNIVERSITY OF
ABERDEEN
The Rowett Institute

n = 15 (B)
gDNA = 9 (B)



Agriculture and
Agri-Food Canada

n = 100 (B)
gDNA = (...)



NUI Galway
OÉ Gaillimh

n = 82 (B); 17 (F)
gDNA = (...)



n = 50 (B); 3 (A)
gDNA = (...)



n = 2 (B)
gDNA = (...)



Instituto Nacional de
Tecnología Agropecuaria

n = 20 (B); 1 (F)
gDNA = (...)



UNIVERSITY OF
ILLINOIS
URBANA-CHAMPAIGN

n = 124 (B)
In stock



南京农业大学
NANJING AGRICULTURAL UNIVERSITY

n = 3 (F)
3 Genomes



UC DAVIS
UNIVERSITY OF CALIFORNIA

n = 2 (F)
gDNA = (2)



Agriculture and
Agri-Food Canada

n = 799 (B), 4 (F)
gDNA = 39 (B)



n = 17180/6087 (B)
2113 Genomes (323-16S)
*146 new species
(≈45%; DRIP 95%, ANI)
(52 media, 17 breed)

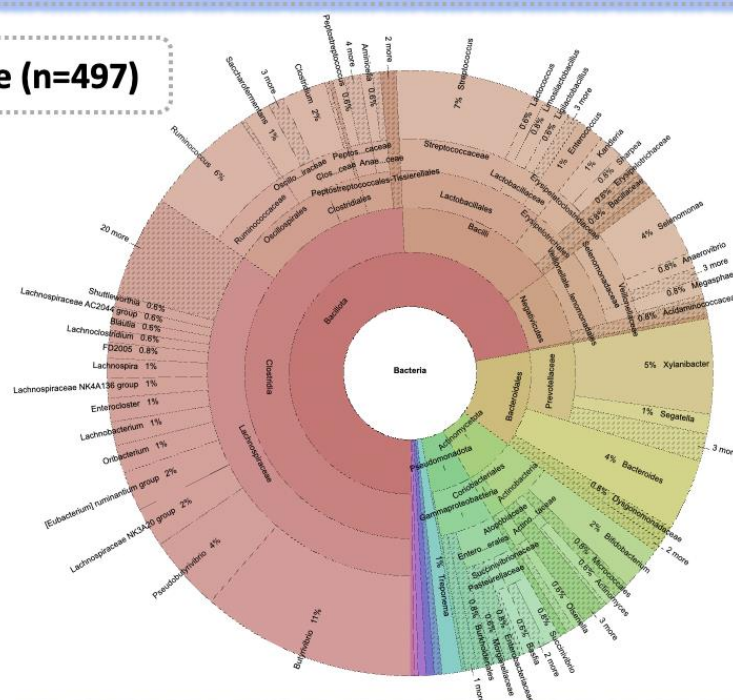
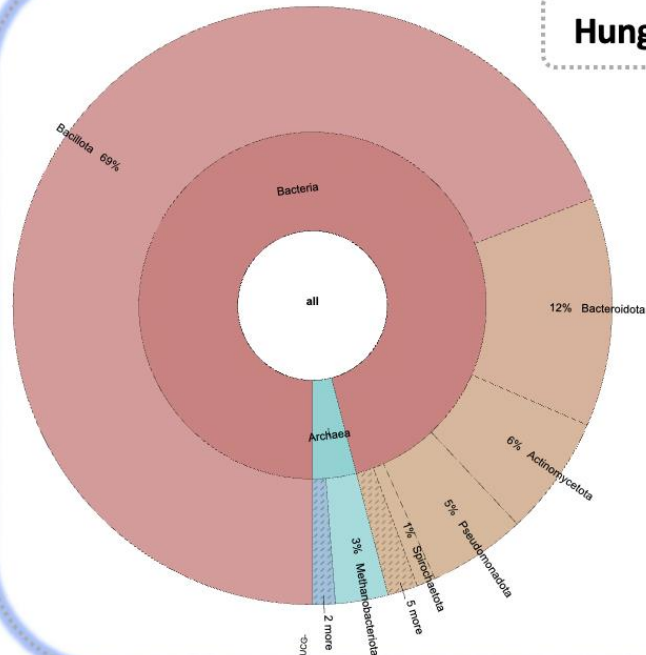


AGROSAVIA
Corporación colombiana de investigación agropecuaria

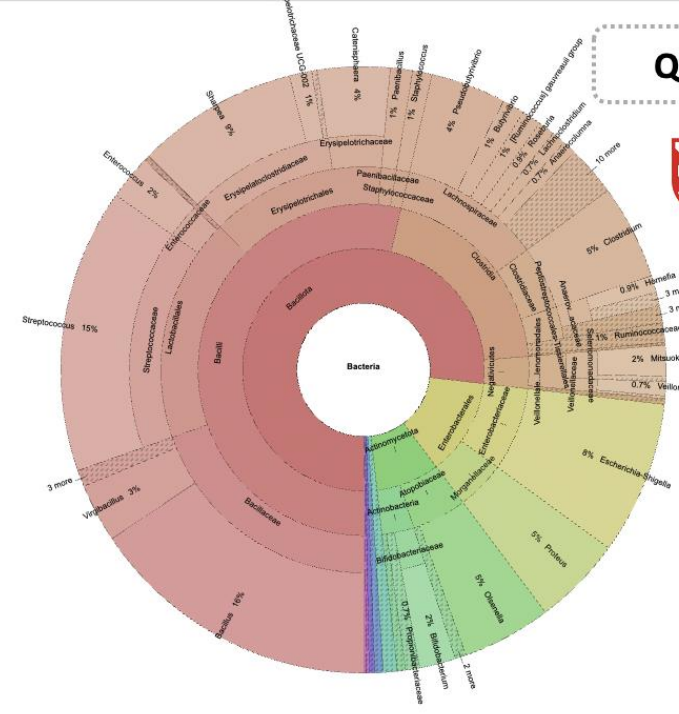
n = 300 (B), 2 (F)
gDNA = 8 (B); 2 (F)

(A) - Archaea; (B) - Bacteria; (F) - Fungi
(...) Initial characterisation for genome
sequencing selection.

Hungate (n=497)



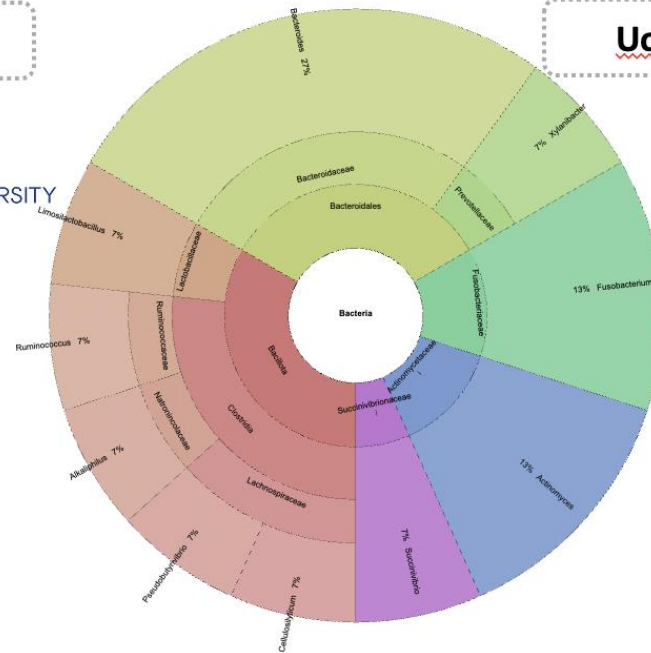
QUB (n=637)



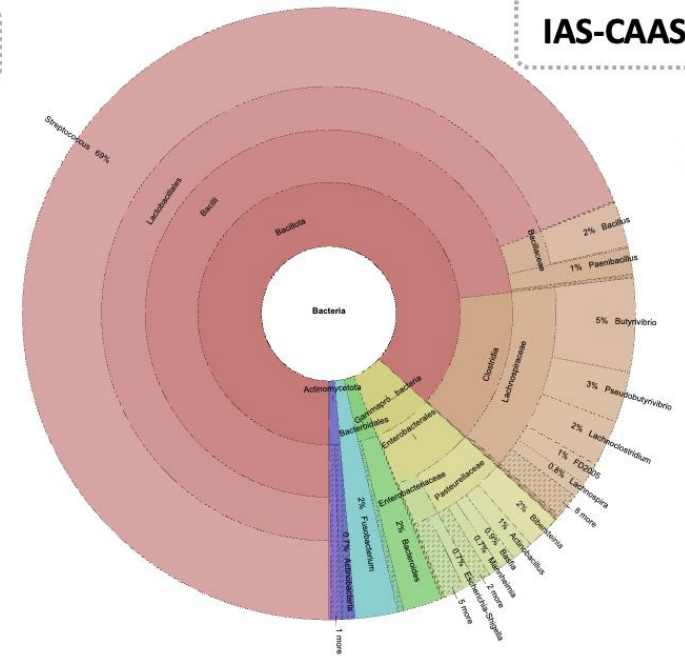
AU (n=38)



UoA (n=15)



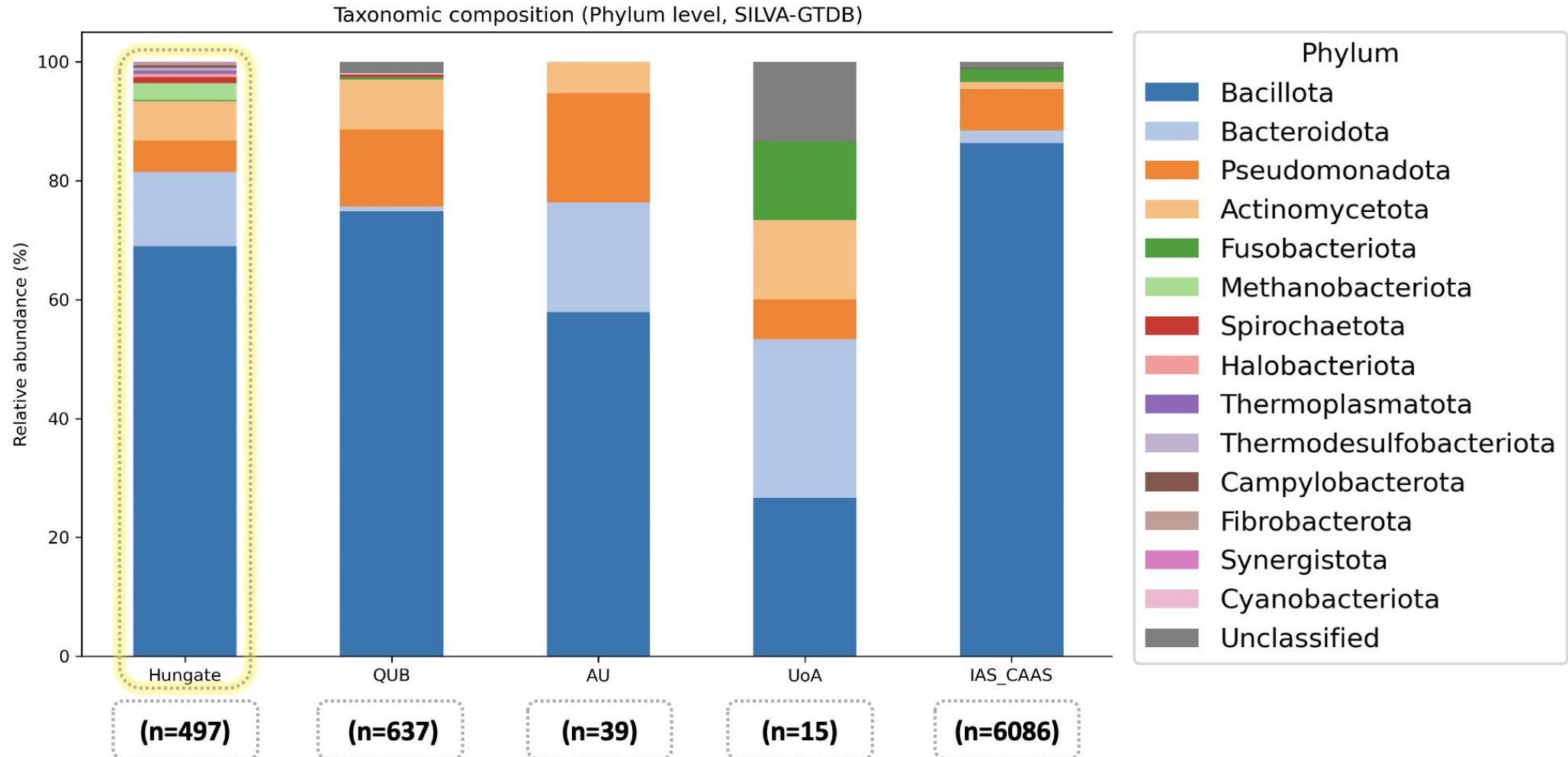
IAS-CAAS (n=6086)



Taxonomic composition at the Phylum level based on partial 16S rRNA gene

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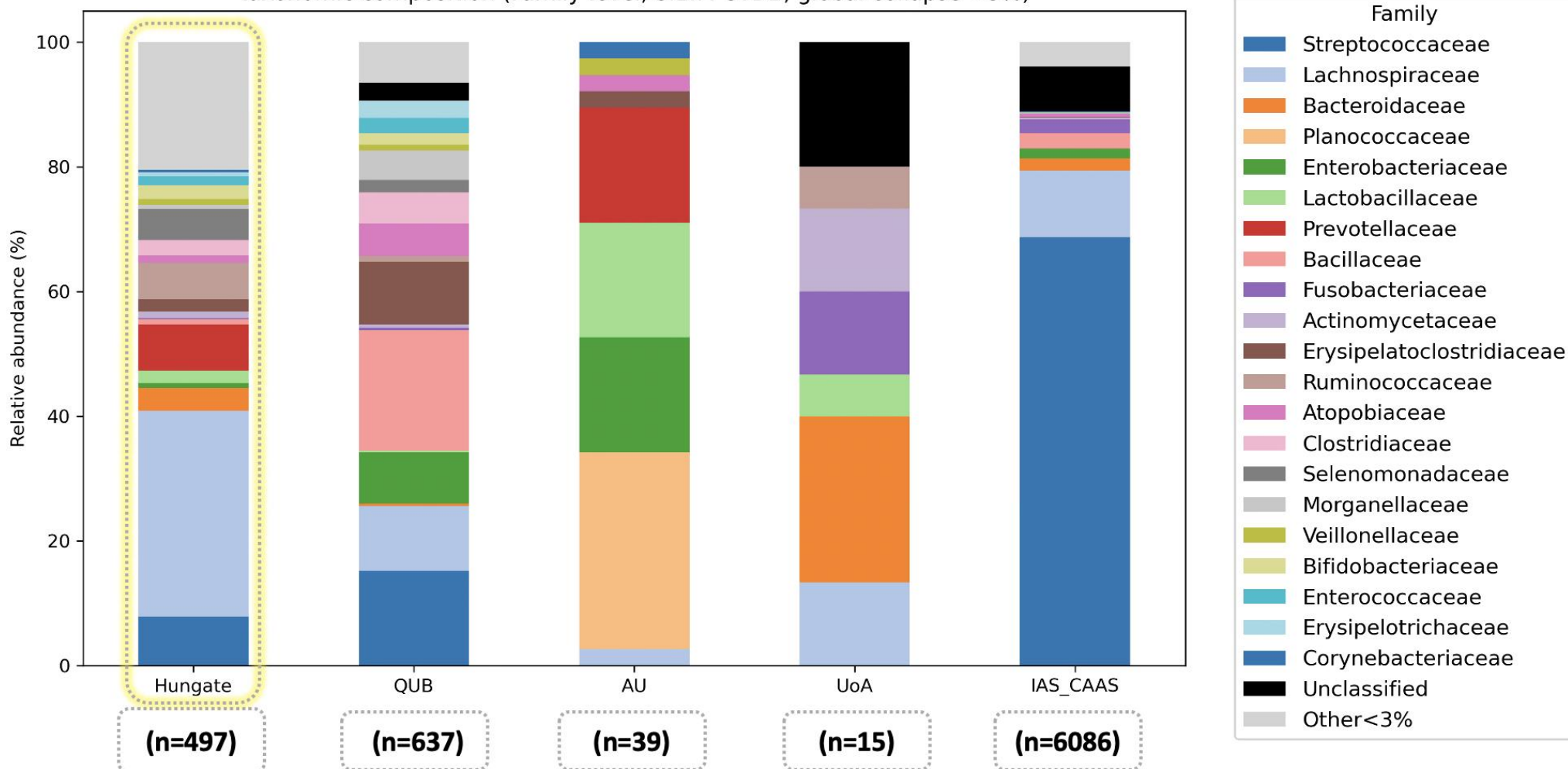


Taxonomic composition at the Family level based on partial 16S rRNA gene

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Taxonomic composition (Family level, SILVA-GTDB, global collapse <3%)



Phylogenetic tree: 16S rRNA gene sequence

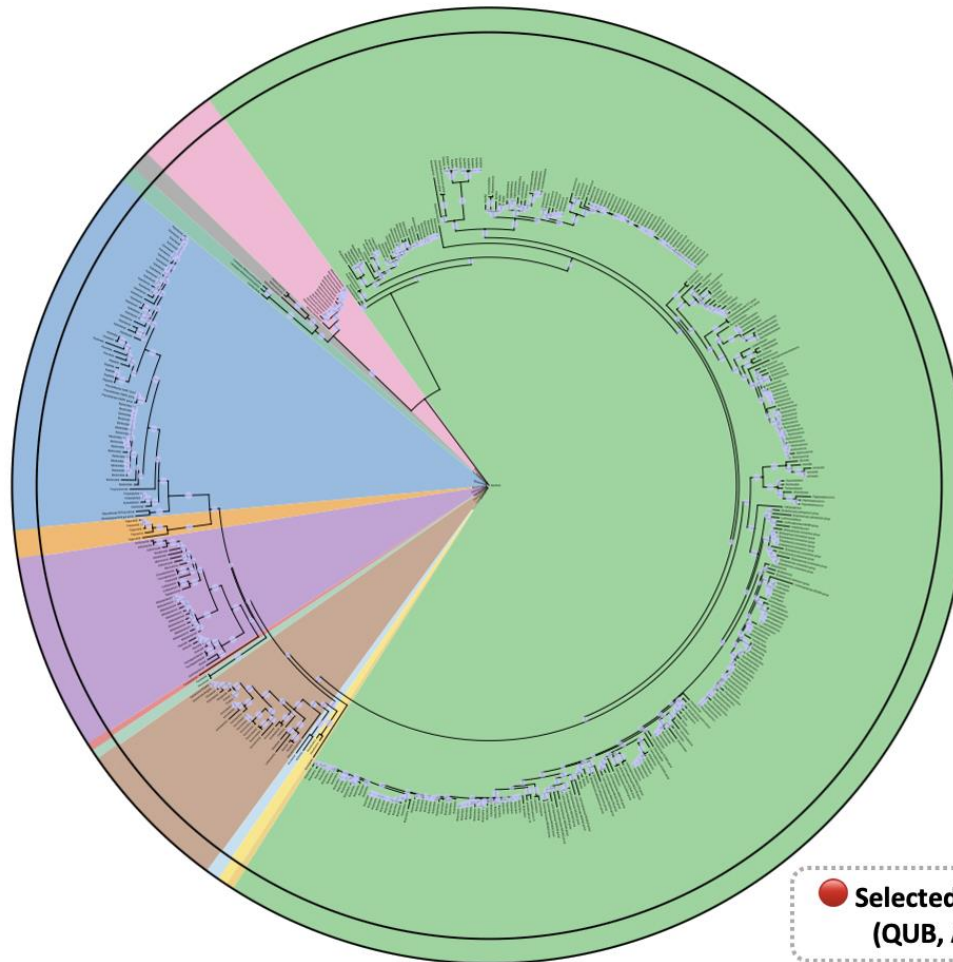
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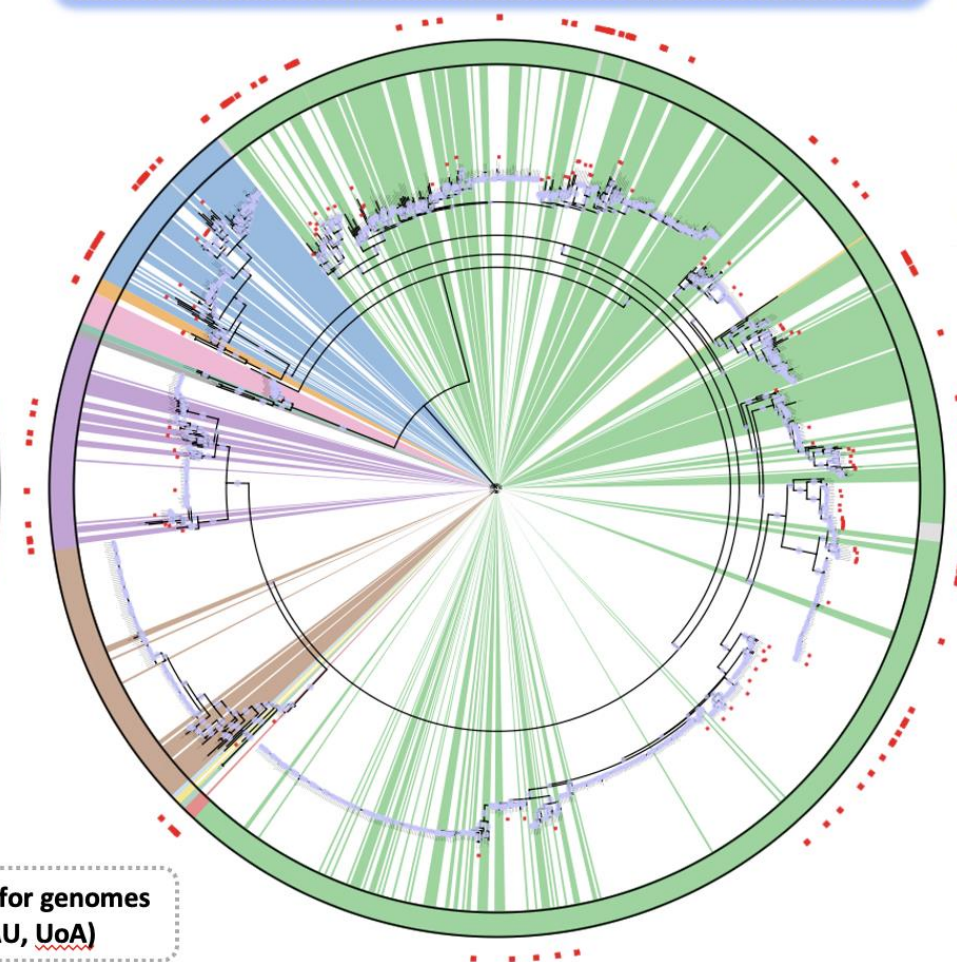
Phylum

- Bacillota
- Actinomycetota
- Bacteroidota
- Spirochaetota
- Fusobacteriota
- Pseudomonadota
- Methanobacteriota
- Halobacteriota
- Fibrobacterota
- Thermodesulfobacteriota
- Campylobacterota
- Synergistota
- Thermoplasmatota

Hungate collection



Hungate collection and new isolates (white background) – B1



bootstrap

- 0
- 25
- 50
- 75
- 100

Selected for genomes
(QUB, AU, UoA)

Phylogenetic tree: 16S rRNA gene sequence

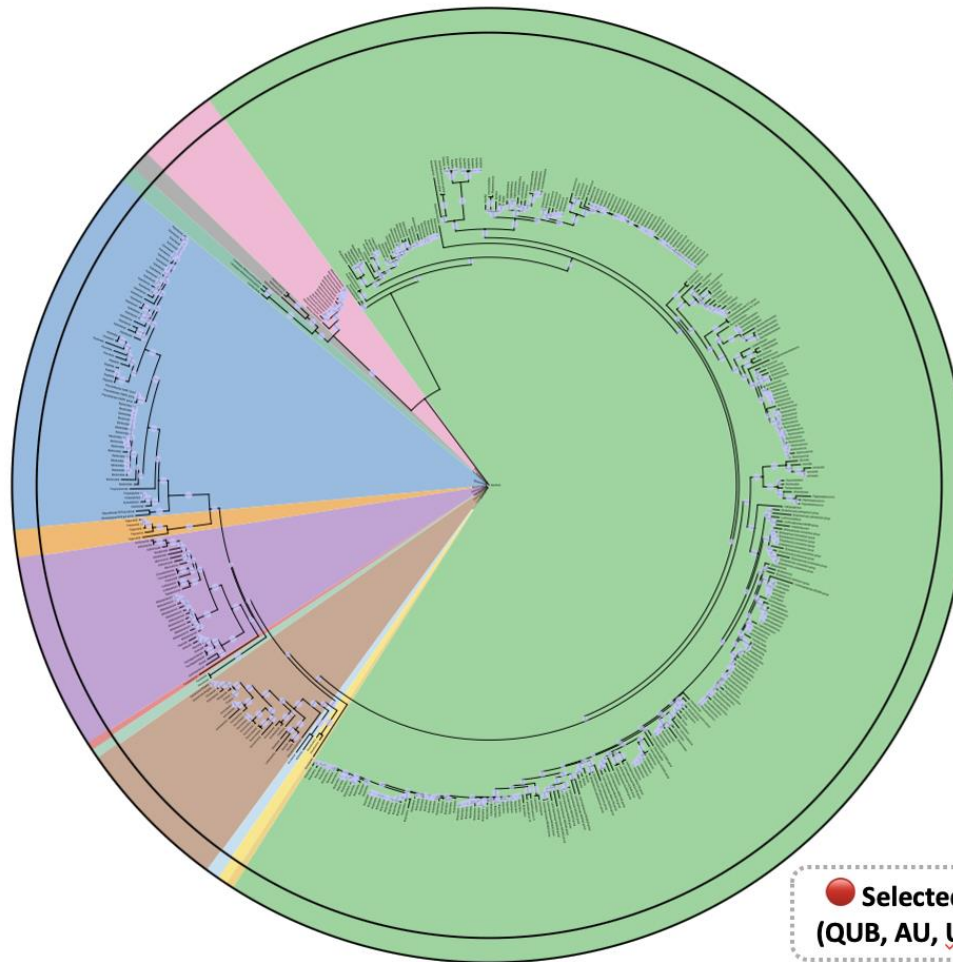
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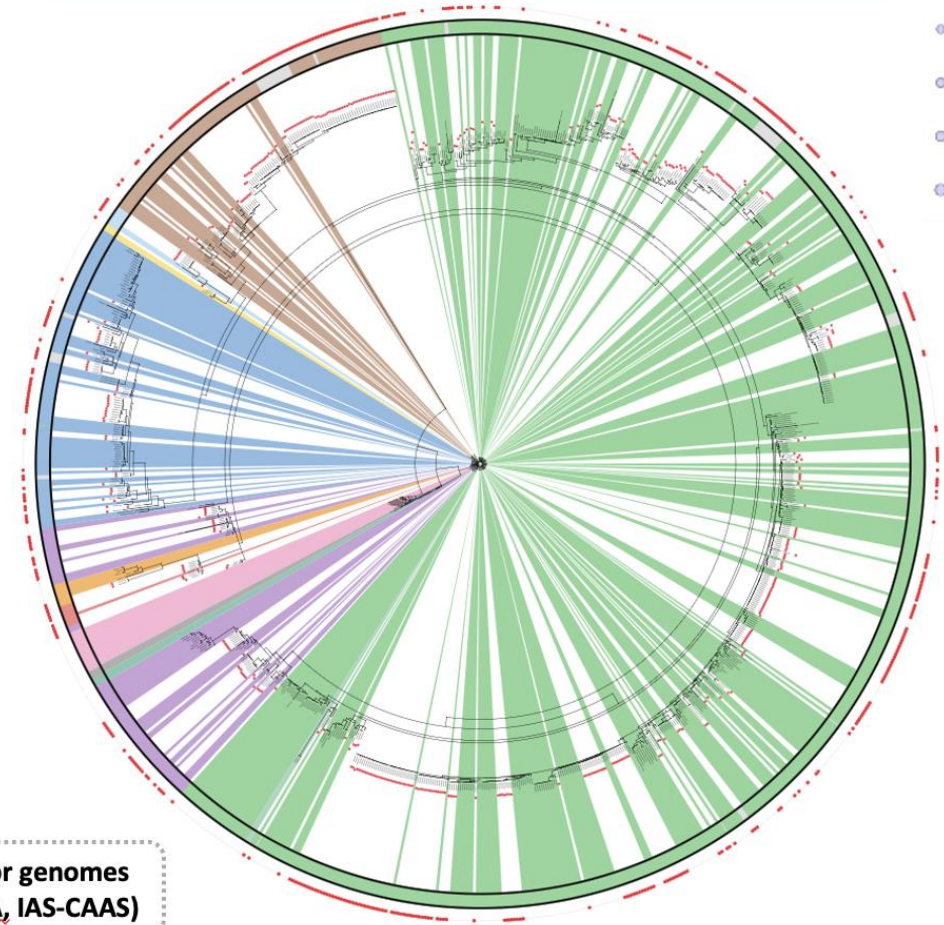
Phylum

- Bacillota
- Actinomycetota
- Bacteroidota
- Spirochaetota
- Fusobacteriota
- Pseudomonadota
- Methanobacteriota
- Halobacteriota
- Fibrobacterota
- Thermodesulfobacteriota
- Campylobacterota
- Synergistota
- Thermoplasmatota

Hungate collection



Hungate collection and new isolates (white background)



bootstrap

- 0
- 25
- 50
- 75
- 100

Selected for genomes
(QUB, AU, UoA, IAS-CAAS)

Thank you for listening and to all the RUMEN Gateway funders and partners!

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1495



An Roinn Talmhaíochta,
Bia agus Mara
Department of Agriculture,
Food and the Marine



NUI Galway
OÉ Gaillimh



AARHUS UNIVERSITY



Agriculture and
Agri-Food Canada



UNIVERSITEIT VAN PRETORIA
UNIVERSITY OF PRETORIA
YUNIBESITHI YA PRETORIA



Instituto Nacional de
Tecnología Agropecuaria



THE UNIVERSITY
OF BRITISH COLUMBIA

BIOMEDIT



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Researchers and collaborators

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Main Hub

QUB: Sharon A. Huws; Fernanda Godoy Santos; James Pickup; Esperanza Fuertes; Idowu Oluwaseun Janet; Katie Lawther; Theano Stoikidou; Ziming Wu; Zhenbin Zhang.

Co-Hub

AAFC: Renee M. Petri; Robert J. Gruninger; Tim A. McAllister; Geneviève Drouin.

AGROSAVIA: Hugo Jiminez; Fernando Rodríguez; Olga Mayorga; Alejandro R. Muñoz (Uniandes)

ARC: Sinalo Mani

AU: Samantha Noel; Anna A. Schönherz; Israt Farhana

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BiomEdit: Arvind Kumar; Dwi Susanti

CSIC: David Yañez-Ruiz; Eva Ramos-Morales

IAS-CAAS: Shengguo Zhao

INRAE: Milka Popova; Diego Morgavi; Arthur Bourguet



INTA: Maria Esperanza Ceron-Cucchi; Silvio Cravero; Fausto Moralejo

NAU: Yanfen Cheng; Weiyun Zhu; Guanghui Hu

RI UoA: Karen P. Scott

UCDavis: Matthias Hess; Timothy John Hackmann

UOG: Sinead M. Waters; Vincent O'Flaherty; Gizem Karakaya; Simon Mills

UIUC: Roderick I. Mackie

UKZN: Matthew A. Adeleke; Peter Anjili Mshelia

UNA: Walter J. Sandoval-Espinola; Mauricio Javier Molinas Vera

UBC: Le Luo Guan

UP: Abiodun M. Akanmu

WUR: Hauke Smidt

In the process of becoming a Co-Hub/sub-contract

CIRDES: Abel S. Biguezoton

EMBRAPA: Pedro Braga Arcuri; Jailton da Costa

Causale: Cauane Blumenberg



