

An overview of the Canadian Collection of Agricultural Soil Microbes (CCASM) and an update on rhizobial taxonomy



BENEFIT

queensu.ca/microbes-for-agriculture

George diCenzo

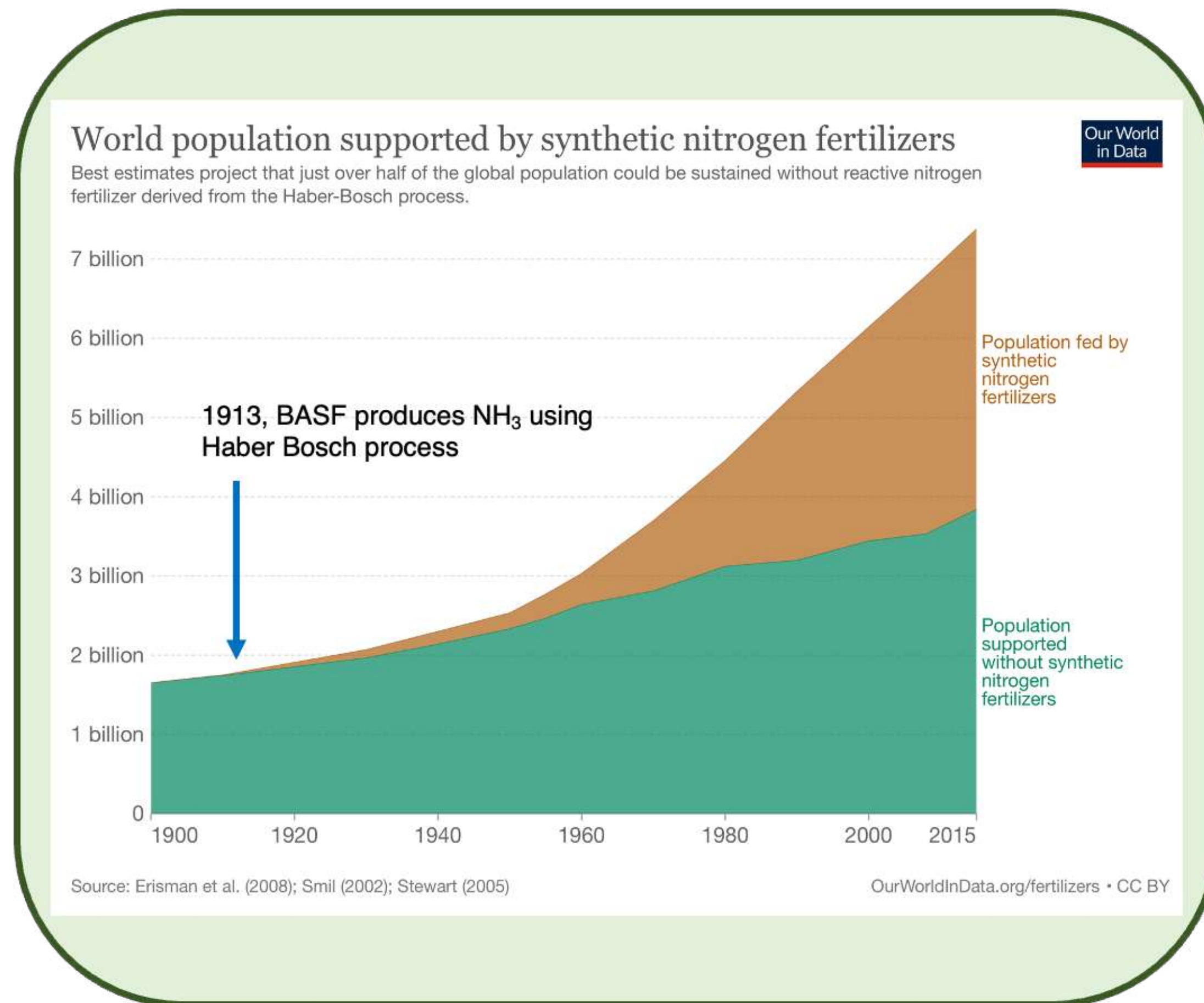
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Impacts of nitrogen fertilizers

Synthetic fertilizers enhance food production



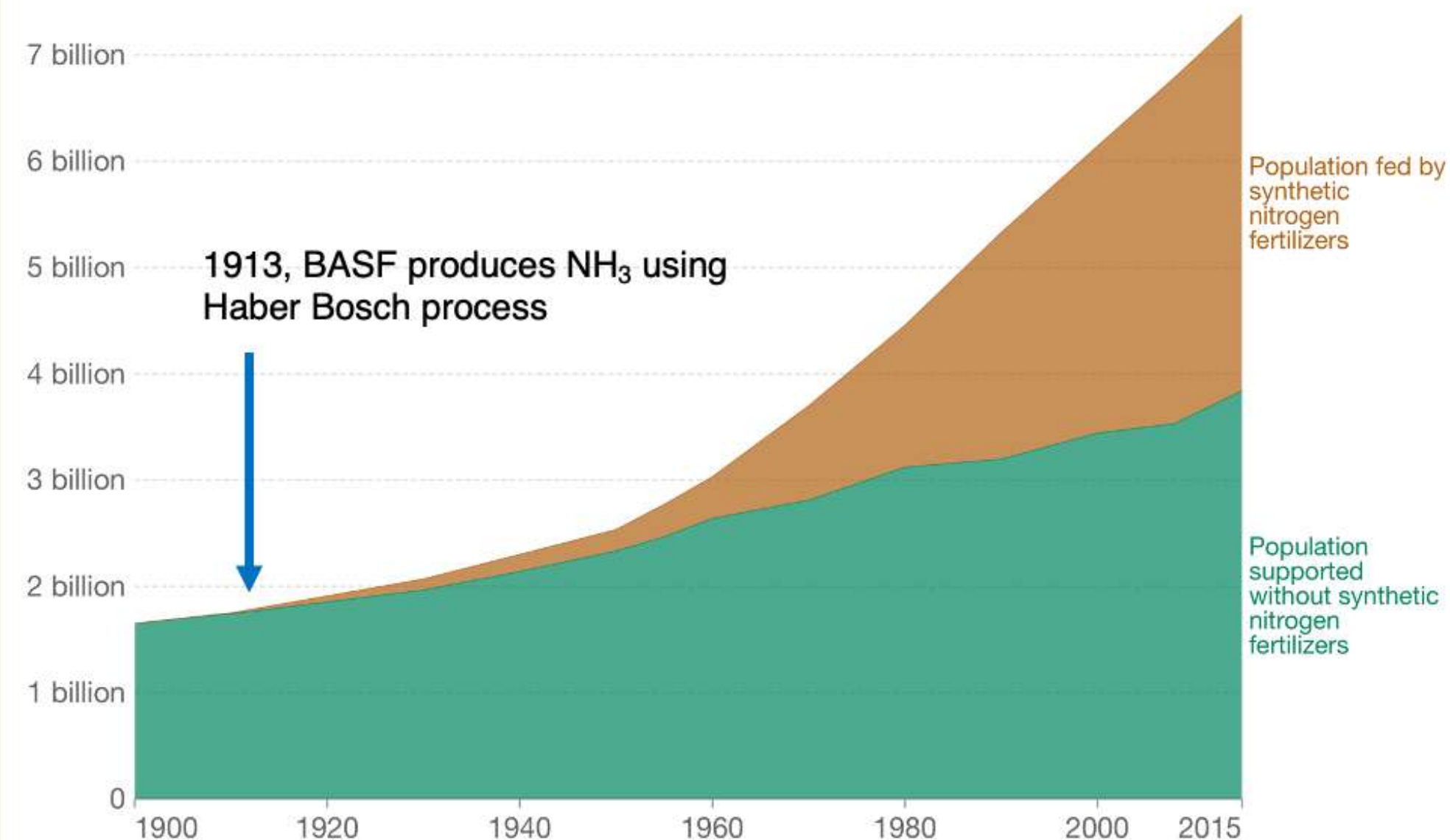
Impacts of nitrogen fertilizers

Synthetic fertilizers enhance food production, but also increase greenhouse gases

World population supported by synthetic nitrogen fertilizers

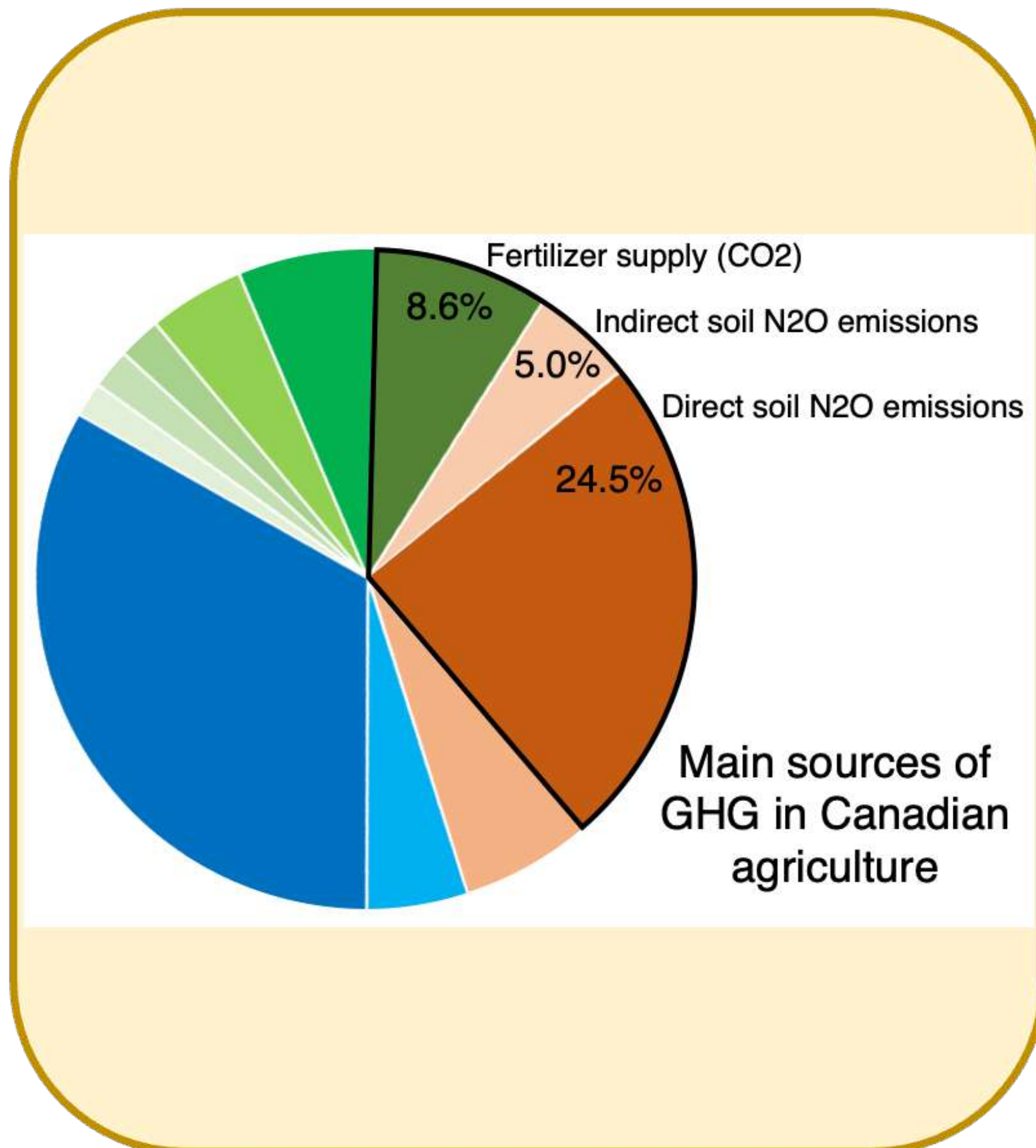
Best estimates project that just over half of the global population could be sustained without reactive nitrogen fertilizer derived from the Haber-Bosch process.

Our World
in Data



Source: Erisman et al. (2008); Smil (2002); Stewart (2005)

OurWorldInData.org/fertilizers • CC BY



The BENEFIT Project

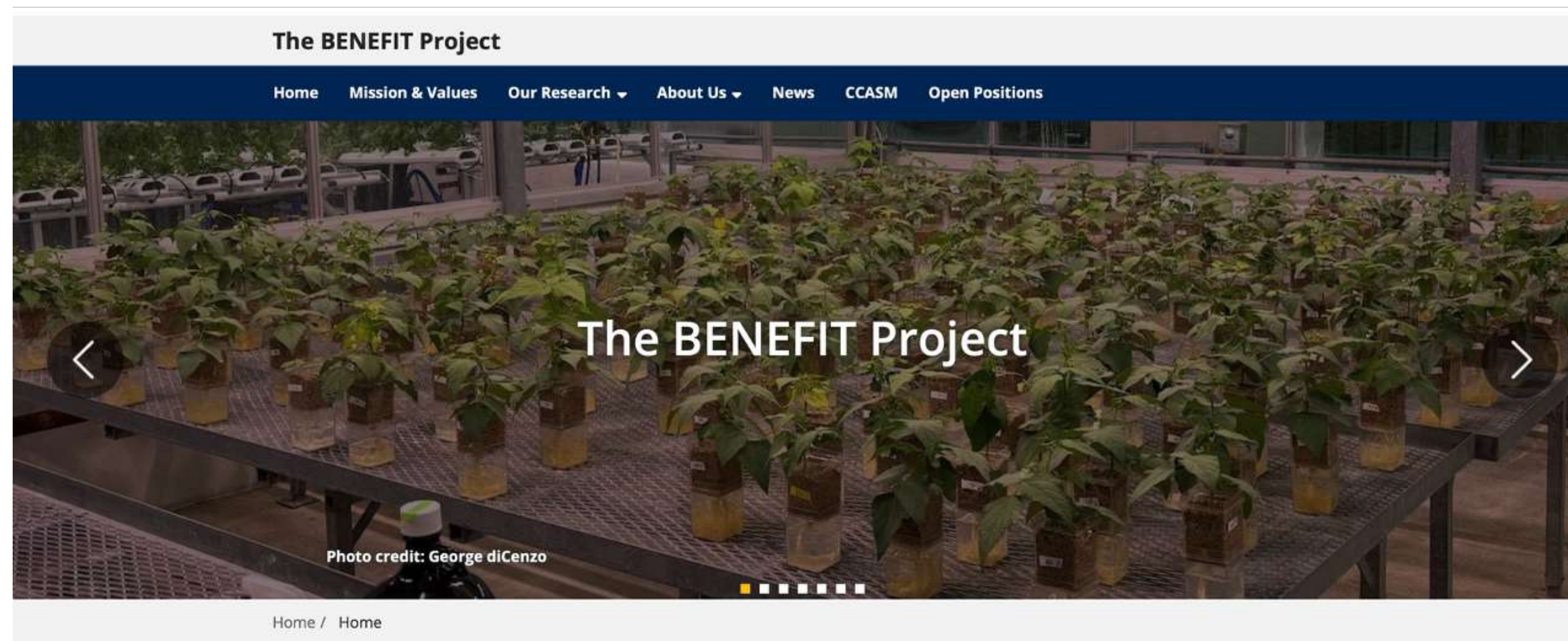


Bio-inoculants for the promotion of nutri**ent** **eff**iciency and crop resiliency in Canadian Agriculture

queensu.ca/microbes-for-agriculture

Identify and optimize microbial inoculants for Canadian crops that allow growers to maintain or increase current crop yields with reduced nutrient input

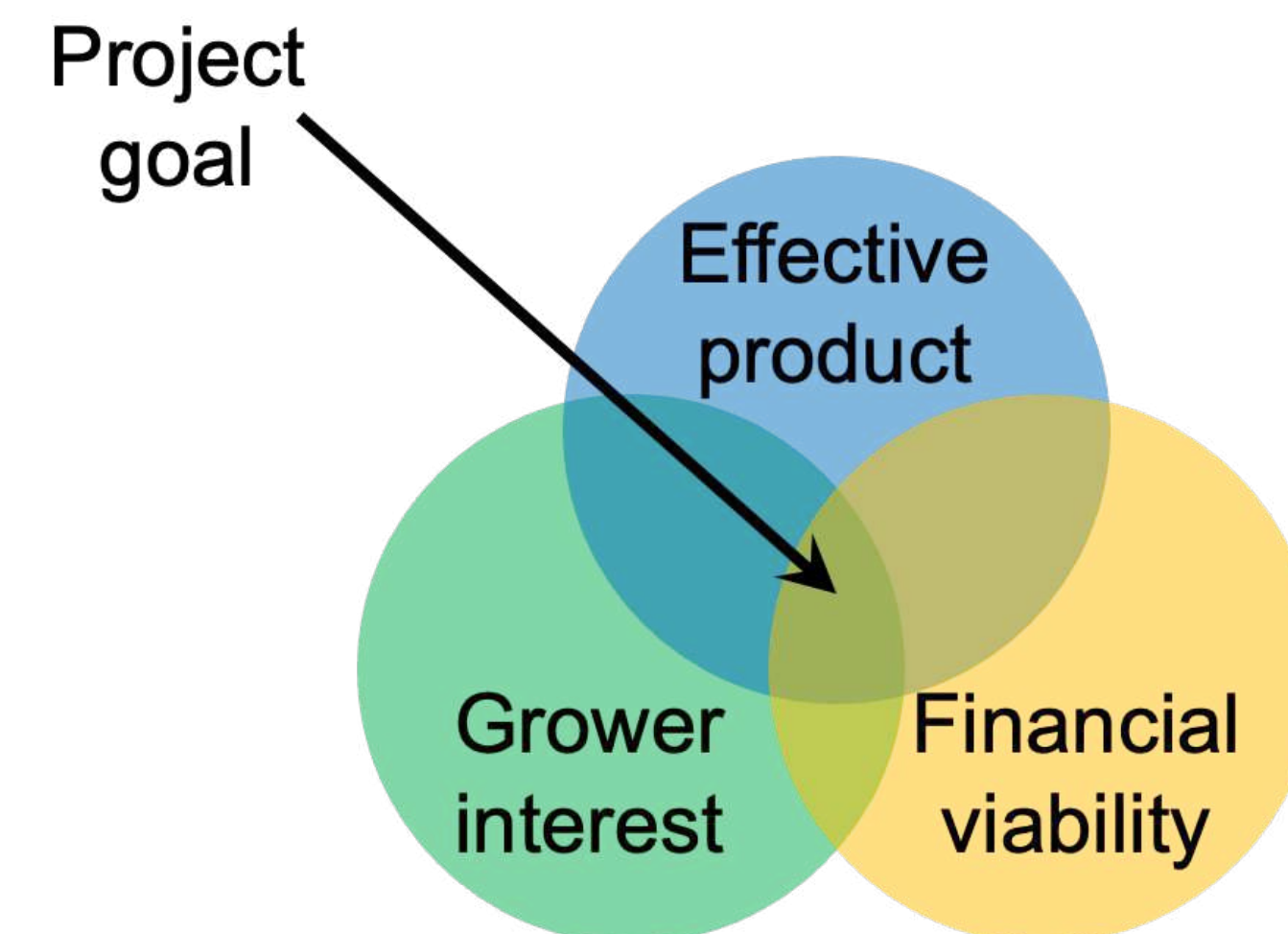
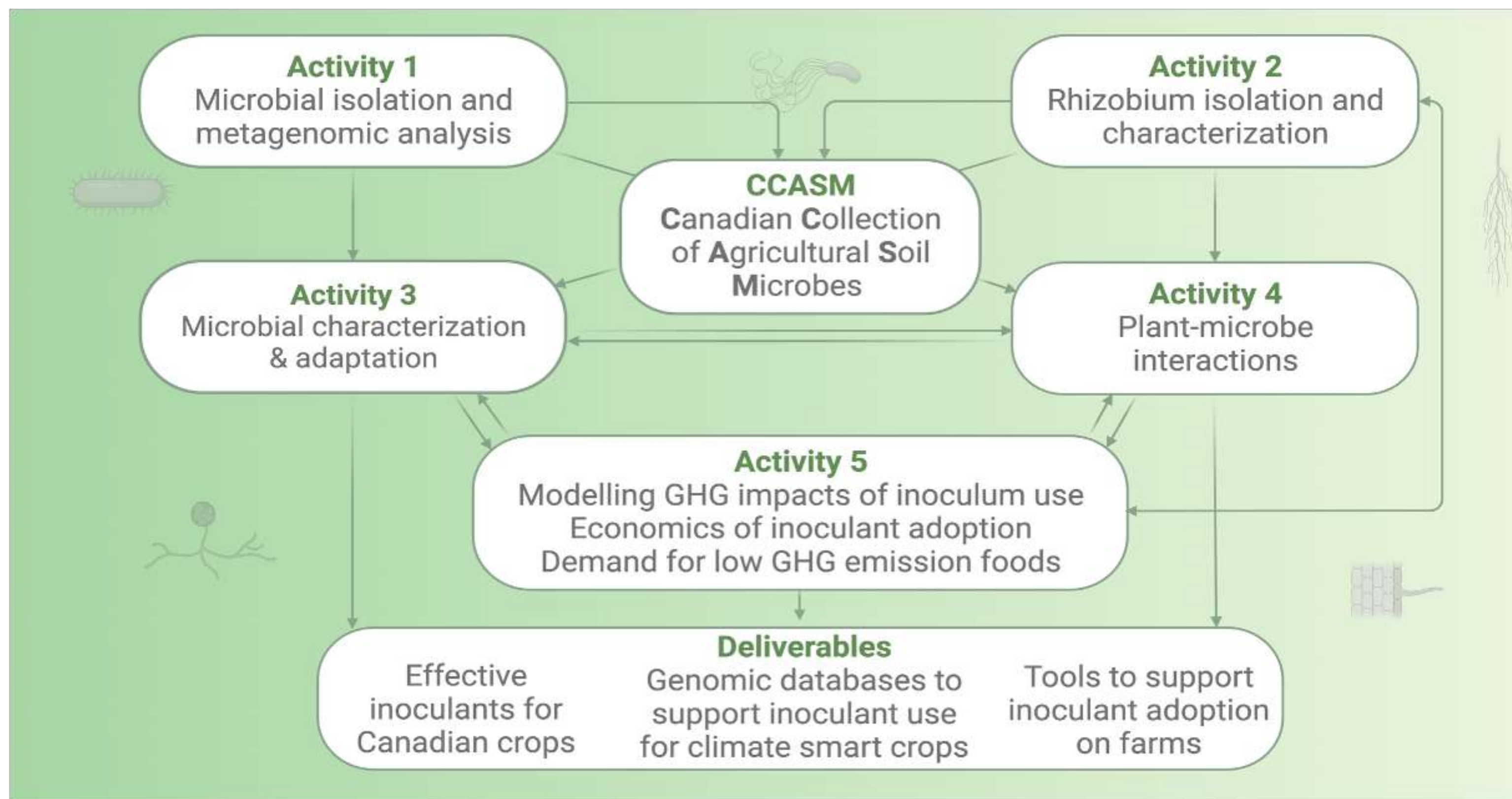
Focused on cereals (wheat, barley), brassicas (canola, kale), and legumes (bean, pea)



The BENEFIT Project



A holistic approach to the development of microbial inoculants, spanning microbe isolation and characterization; elucidation of plant responses; and the economic and environmental impact.



Developing new inoculants for Canada



Where do we start?

First, need a bio-bank of bacterial isolates to screen for traits of interest

Prioritize isolates from Canadian soils

A diverse library of agricultural soil bacteria



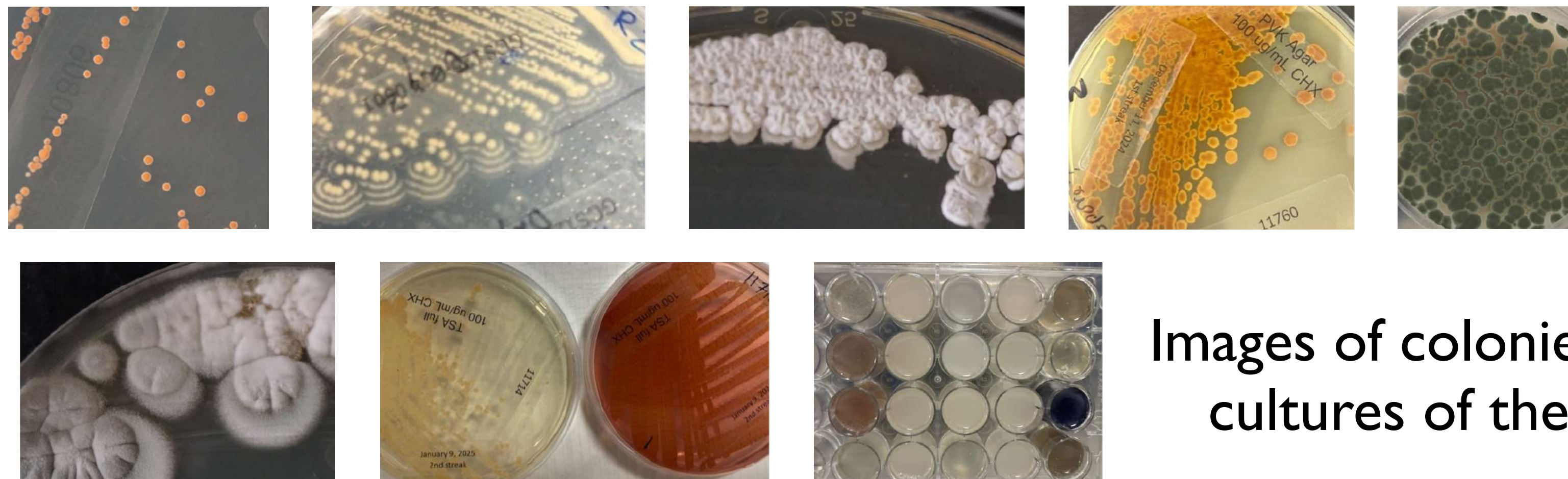
We have collected over 2,000 bacterial isolates from soils collected across Ontario and the Prairies and completed WGS of all isolates

Of ~1,100 Ontario isolates

≥288 species from 88 genera across 38 families in 4 phyla

Bacillota (57%), *Actinomycetota* (30%), *Pseudomonadota* (12%), *Bacteroidota* (1%)

Potentially ≥150 putatively undescribed species



Images of colonies or liquid cultures of the isolates

A collection of legume nodule isolates

We have collected and genome-sequenced over 1,200 bacterial isolates from legume nodules (common bean, pea, adzuki bean, and soybean) using soils from Ontario and Manitoba



Navy bean plants inoculated with rhizobia isolated from Ontario soils

A collection of bacteria from arctic soils



We have collected and genome-sequenced 99 bacterial isolates (19 species) from near Daring Lake, Northwest Territories



Questions arising from this sampling effort



Why go through the trouble of generating our own collection?

What do we do with all of these isolates?

Why generate our own bacterial collection?



Comment from a reviewer: “How this differs or complements previous scouting work for microbial inoculants in Canadian agriculture would help the panel better understand the need.”

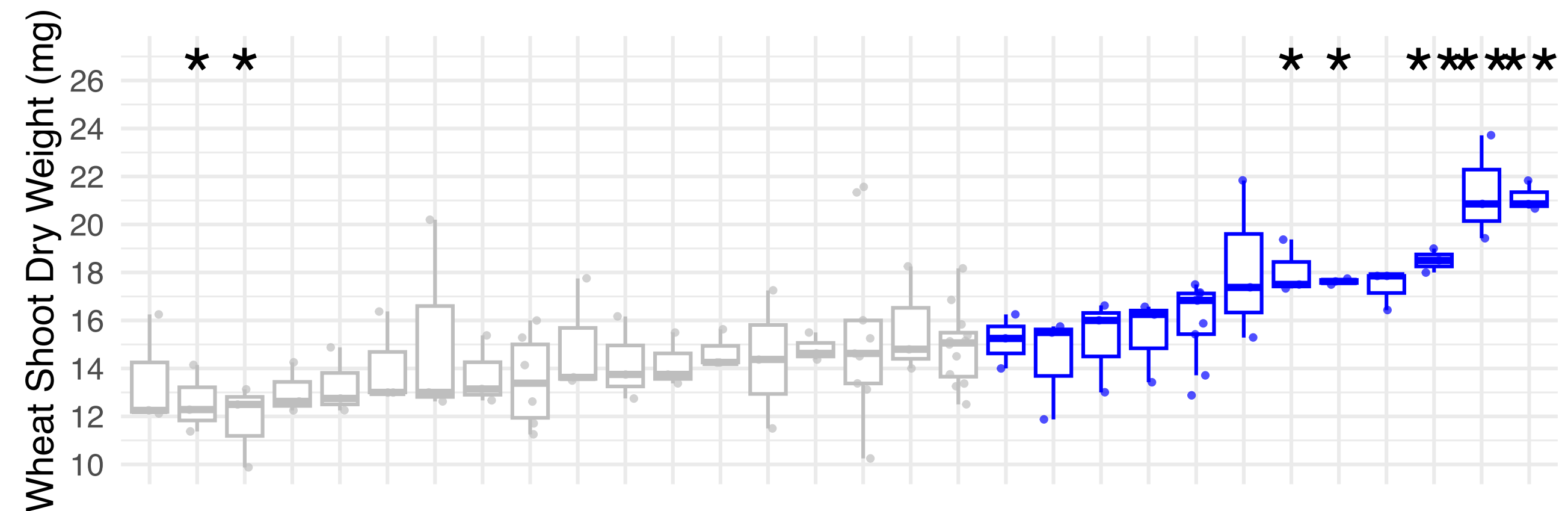
Response: “...Collections of Canadian strains from other studies are not readily available and so we must first establish our own collection...”

What do we do with all these isolates?



Screen for traits relevant to bio-inoculant success

Plant growth promotion



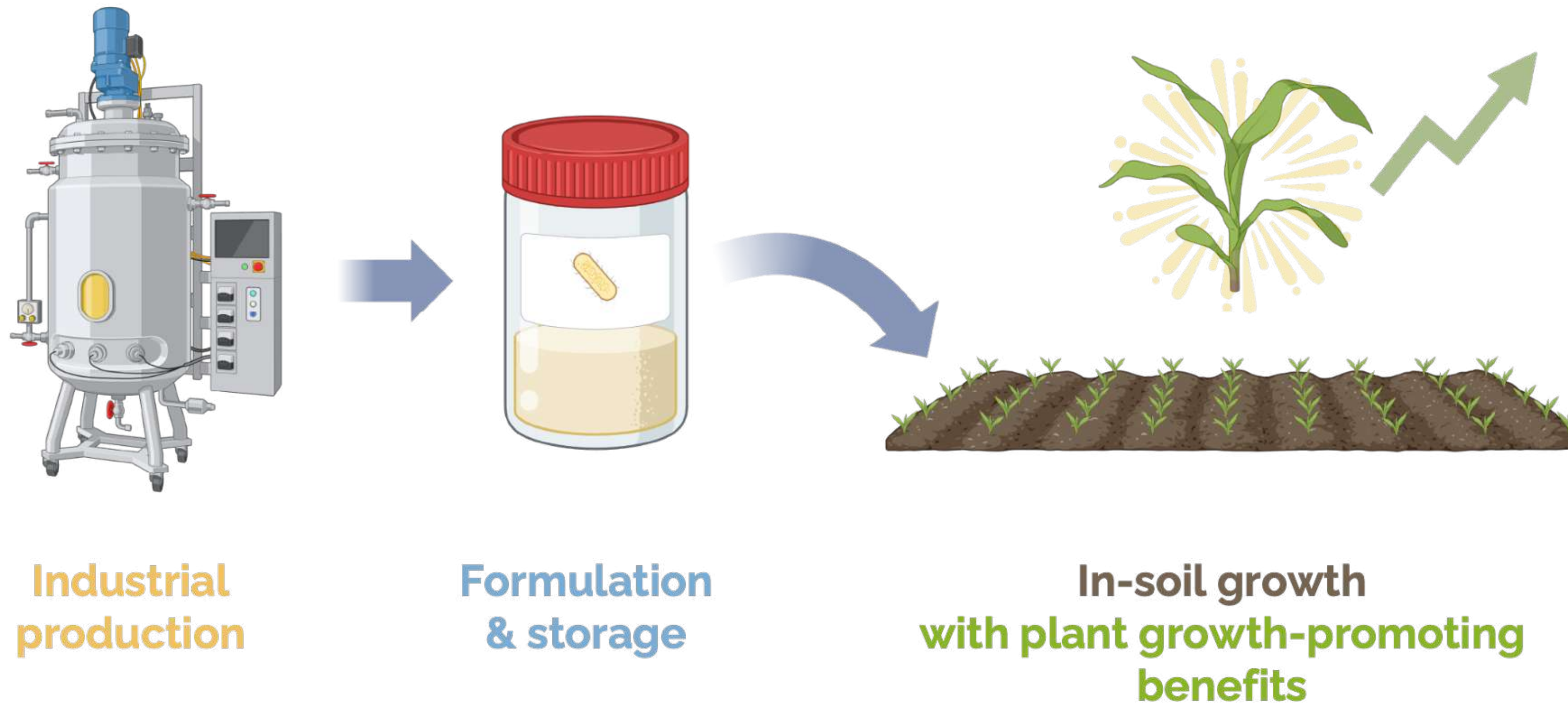
Impacts of isolates on the weight of wheat seedlings

What do we do with all these isolates?

Screen for traits relevant to bio-inoculant success

Plant growth promotion

Robust across industry pipeline (Hemara et al. 2026. ISME Communications. ycag190)



What do we do with all these isolates?

Screen for traits relevant to bio-inoculant success

- Plant growth promotion

- Robust across industry pipeline (Hemara et al. 2026. ISME Communications. ycag190)

But there are many other potential uses of the isolates!

- Examine ability to protect crops from abiotic stress

- Evaluate bio-control potential

- Screen for other traits relevant to biotechnology

- Screen for novel antimicrobial compounds

- Characterization of novel species

Thus important that the isolates be accessible for reuse by others

Canadian Collection of Agricultural Soil Microbes (CCASM)



A new culture collection designed to facilitate the sharing of bacteria (and associated genomic data) isolated from agricultural soils within Canada

Duplicated at Queen's University and the University of Manitoba (Dr. Matthew Bakker)

Stored as glycerol stocks at -80°C in barcoded vials



Canadian Collection of Agricultural Soil Microbes

Canadian Collection of Agricultural Soil Microbes (CCASM)



- Will contain all isolates collected as part of BENEFIT
- Will accept deposits from other research groups
- Distribution and deposits initially limited to just Canada
- A nominal fee associated with use
- Not yet public but should be public this year



Canadian Collection of Agricultural Soil Microbes

Canadian Collection of Agricultural Soil Microbes (CCASM)



Home page provides a geographic breakdown of the source of isolates

 Canadian Collection of Agricultural Soil Microbes

[Browse](#) | [Statistics](#) | [Deposit](#) | [Cart](#)



Welcome to the Canadian Collection of Agricultural Soil Microbes

The Canadian Collection of Agricultural Soil Microbes (CCASM) is a collection of bacteria isolated primarily from agricultural fields across Canada. CCASM is hosted by Queen's University and the University of Manitoba, which was designed to facilitate the sharing of agricultural soil microbes across Canada.

Browse the Collection

CCASM is hosted at Queen's University and the University of Manitoba. Initial development of CCASM was supported by funding from Genome Canada, the Government of Manitoba, and the Government of Ontario.



Strains per Province:

Strains from British Columbia: 4
Strains from Alberta: 2
Strains from Saskatchewan: 0
Strains from Manitoba: 0
Strains from Ontario: 8
Strains from Quebec: 1
Strains from Newfoundland and Labrador: 0
Strains from New Brunswick: 0
Strains from Prince Edward Island: 0
Strains from Nova Scotia: 0
Strains from Yukon: 0
Strains from Northwest Territories: 0
Strains from Nunavut: 0

Total in Collection: 15

Go To Statistics

[About](#) | [Admin](#)

Canadian Collection of Agricultural Soil Microbes (CCASM)



Can browse the collection based on various parameters

 Canadian Collection of Agricultural Soil Microbes

[Browse](#) | [Statistics](#) | [Deposit](#) | [Cart](#)

Browse the Canadian Collection of Agricultural Soil Microbes (CCASM)

This page can be used to search CCASM. The simple search will search just the binomial name (i.e., genus and species) field of each entry. The complex search can be used to additionally search the following fields: full taxonomic lineage, CCASM ID, strain name, associated plant species, and province of isolation. The binomical name, associated plant species, and province of isolation search fields include an optional dropdown menu. Upon selecting a strain of interest, there is an option to add the strain to the cart to facilitate ordering of the strain.

☐ Simple ☒ Complex

Binomial Classification
Rhizobium anhuientes

Full Taxonomic Lineage

CCASM ID

Strain Name

Associated Plant Species

Province

Search

Clear Results

CCASM ID: CCASM_456456 Binomial Classification: Rhizobium anhuientes
Taxonomic Lineage: Bacteria; Pseudomonadota; Alphaproteobacteria; Hyphomicrobiales; Rhizobiaceae; Rhizobium; Rhizobium anhuiese

CCASM ID: CCASM_420420 Binomial Classification: Rhizobium anhuientes
Taxonomic Lineage: Bacteria; Pseudomonadota; Alphaproteobacteria; Hyphomicrobiales; Rhizobiaceae; Rhizobium; Rhizobium anhuiese

CCASM ID: CCASM_010101 Binomial Classification: Rhizobium anhuientes
Taxonomic Lineage: Bacteria; Pseudomonadota; Alphaproteobacteria; Hyphomicrobiales; Rhizobiaceae; Rhizobium; Rhizobium anhuiese

Items per page: 5 1 – 3 of 3

[About](#) | [Admin](#)

Canadian Collection of Agricultural Soil Microbes (CCASM)



Selecting a strain provides various pieces of information, including soil information (if available), genome accessions, and optionally a photo

The screenshot shows the CCASM website interface. A modal window titled "Strain Information" is open, displaying the following details for strain CCASM_456456:

- CCASM ID:** CCASM_456456
- Strain Name:** QUR0207b
- Binomial Classification:** *Rhizobium anhuienses*
- Taxonomic Lineage:** Bacteria; Pseudomonadota; Alphaproteobacteria; Hyphomicrobiales; Rhizobiaceae; Rhizobium; Rhizobium anhuiense
- Risk Group:** 1
- Is Plant Pathogen:** false
- Colony Morphology:** Pale and mucoidy
- Province of Isolation:** BC
- Associated Plant Species:** *Phaseolus vulgaris*
- Isolation Source:** Nodule
- Isolation Year:** 2023
- Isolation Protocol:** Nodule trapping
- Isolation Growth Temperature (°C):** 28
- Isolation Growth Medium:** TY
- Isolation Growth Medium Composition:** 5 g/VL tryptone, 2.5 g/VL yeast extract, 10 mM CaCl₂
- Isolation Soil pH:** 6.5
- Isolation Soil Organic Matter:** 4
- Isolation Soil Texture:** Loam
- Genome NCBI Association:** BioProject XXX, SRA YYY
- Genome Size:** 7295560
- Notes:** N/A

The background shows a search results page with filters for "Simple" and "Complex" search, a "Binomial Classification" filter set to *Rhizobium anhuienses*, and a "Province" filter. A list of search results is visible, including CCASM IDs and taxonomic lineages. The modal window has "Cancel" and "Add to Cart" buttons at the bottom right.

All information in these screenshots is based on test data

Canadian Collection of Agricultural Soil Microbes (CCASM)



Can submit forms to initiate deposit or ordering of strains

DEPOSIT STRAIN FORM

If you are interested in depositing your isolate(s) in CCASM, please fill-in and submit this form to initiate the process.

First name*	Last name*	Email*
Affiliation*		
Description of isolates*		

I have read and agree to the [Terms and Conditions](#)

- ☐ By submitting this form, you consent to receiving an email from the collection curators, asking for more information and who will provide instructions on how to submit your isolate(s). Submission of this form does not guarantee that your isolates will be accepted for deposit in CCASM; only strains meeting the requirements (e.g., isolated from a Canadian source) will be accepted.

[✓ Submit](#) [↓ Get Template CSV](#)

REQUEST STRAIN FORM

To request one or more isolates be shipped to you, please add the strains of interest to the cart, and then fill-in and submit this form to initiate the process.

First name*	Last name*	Email*
Affiliation*		
Additional Information		

I have read and agree to the [Terms and Conditions](#)

- ☐ By submitting this form, you consent to receiving an email from the collection curators, asking for more information and who will provide instructions on how to receive your isolate(s). Submission of this form does not guarantee that your request will be fulfilled; only requests meeting the requirements (e.g., shipping to an address in Canada) will be fulfilled.

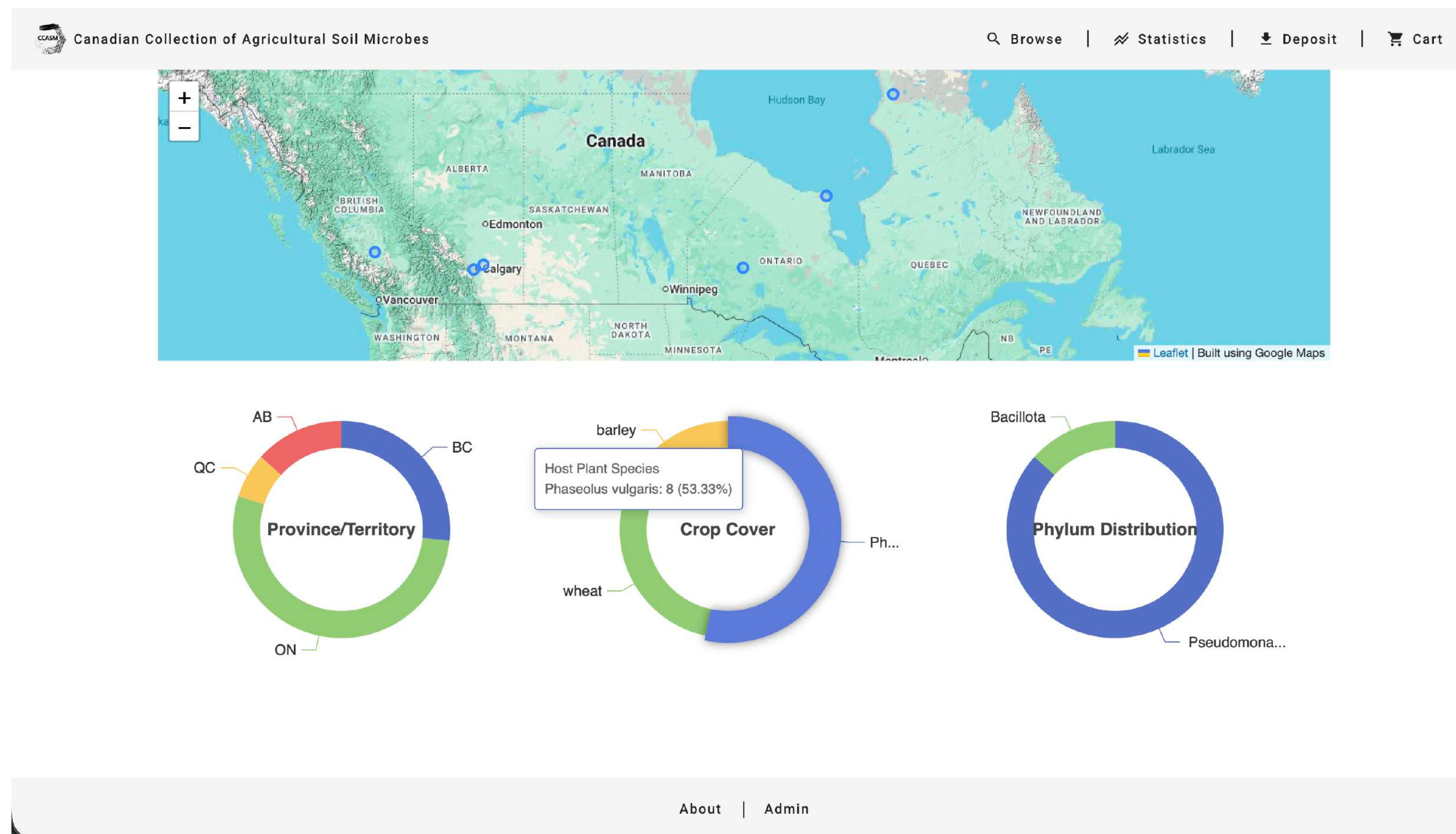
* You don't have anything in cart...

[✓ Submit](#) [↓ Save Metadata](#)

Canadian Collection of Agricultural Soil Microbes (CCASM)



Statistics page provides some high-level statistics summarizing the collection



Canadian Collection of Agricultural Soil Microbes (CCASM)



A new culture collection designed to facilitate the sharing of bacteria (and associated genomic data) isolated from agricultural soils within Canada

Support reuse of isolates, open distribution of strains and metadata, and reduce the need for new sampling efforts



Introduction to rhizobia and their taxonomy

Rhizobium - legume symbioses

Legumes associate with nitrogen-fixing bacteria called rhizobia that colonize a legume organ known as a nodule



Left - rhizobia
Right - no rhizobia



Dark - rhizobia
Light - no rhizobia



Left - rhizobia
Right - no rhizobia

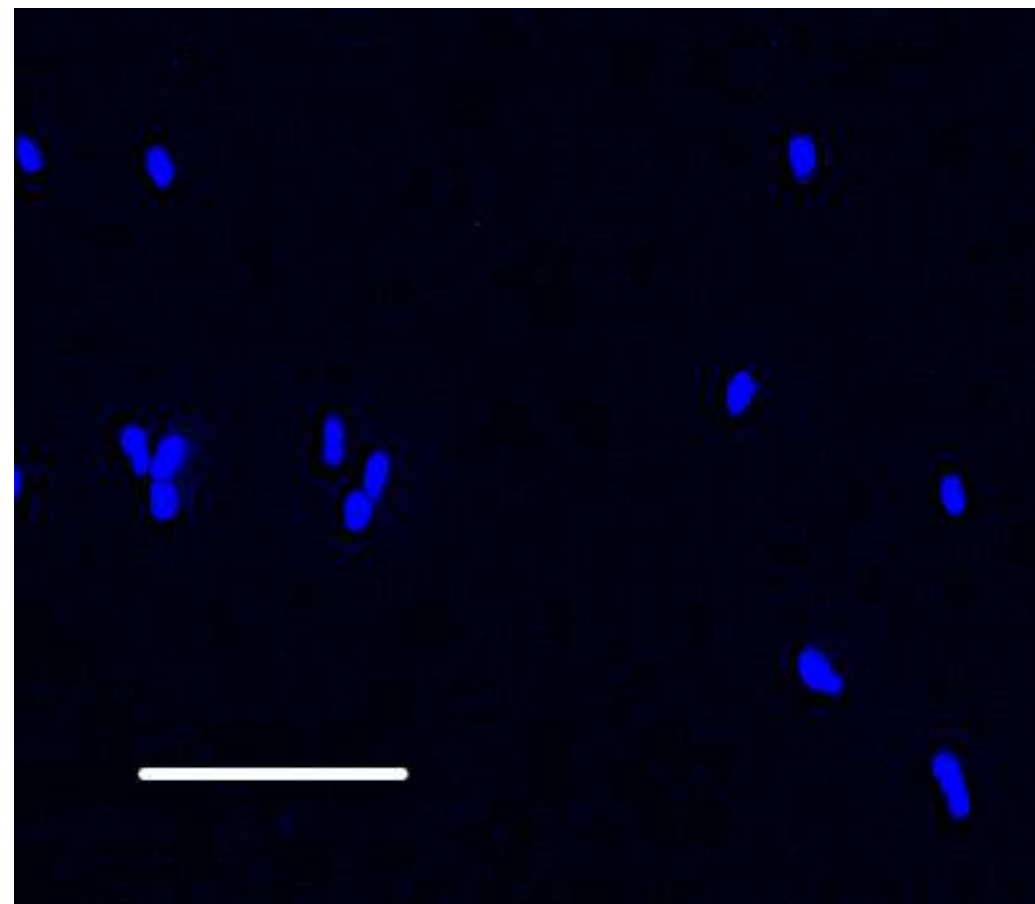
Rhizobium - legume symbioses

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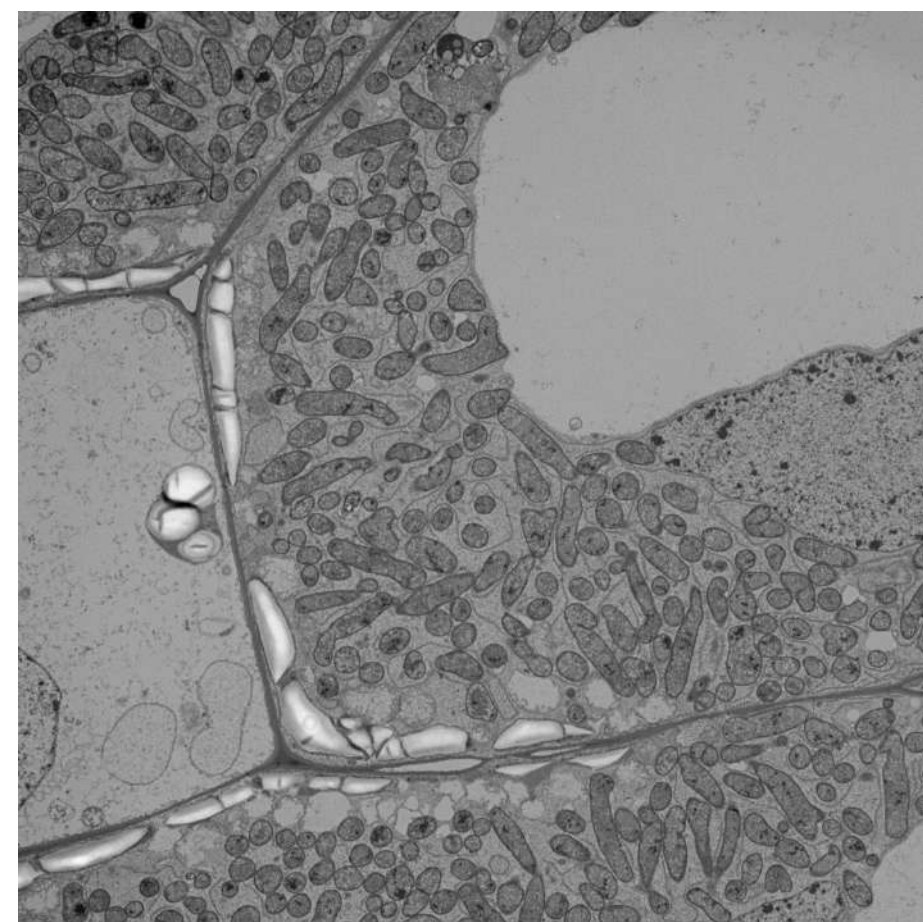
Alfalfa root
nodule



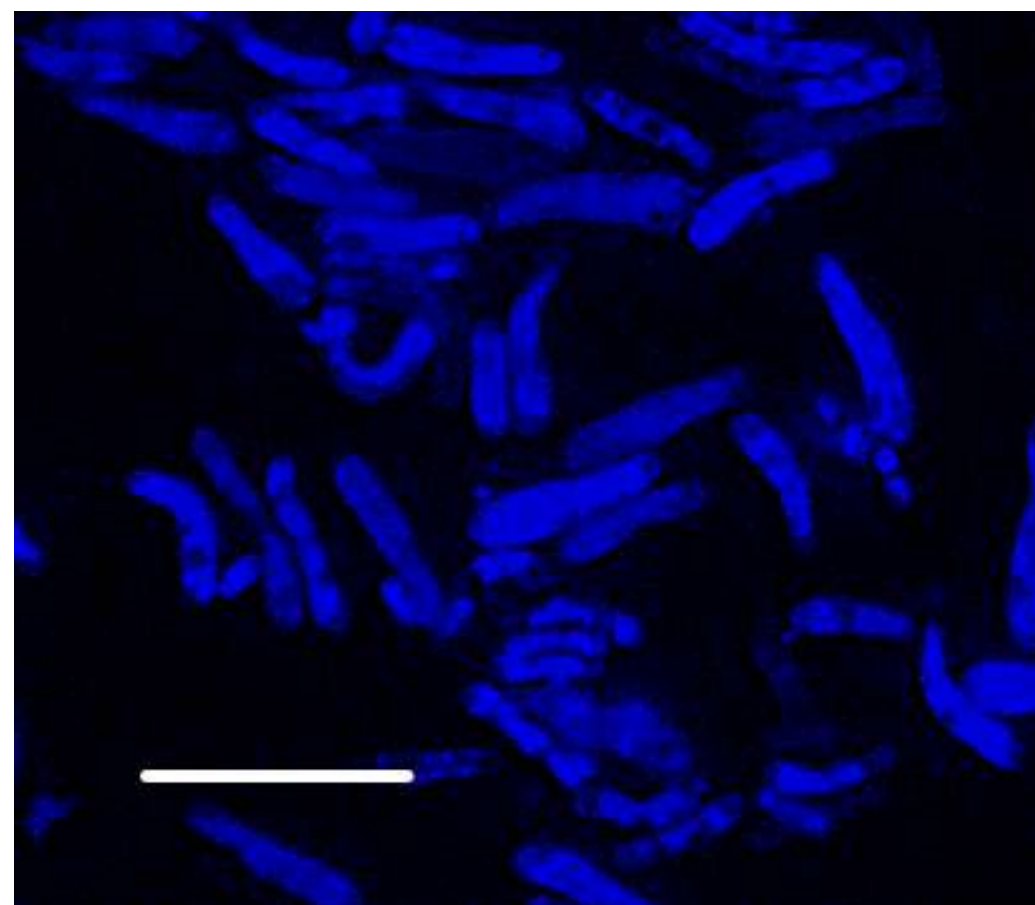
Free-living *S. meliloti*



TEM image of a nodule
cross-section



S. meliloti bacteroids



Tree scale: 0.1

The following table lists the bacterial strains shown in the phylogenetic tree, grouped by their approximate position from top to bottom:

- Pyruvibacter mobilis* CGMCC 1.15125 T
- Tepidicaulis marinus* MA2 T
- Parvibaculum lavamentivorans* DS-1 T
- Rhodogotrophus appendicifer* 120-1 T
- Aestuariivirga litorealis* KC7C 52945 T
- Methyloceanibacter caenitipici* Gela T
- Hypnomicrobium denitrificans* ATCC 51888 T
- Rhodomicrobium vannielii* ATCC 17100 T
- Dichotomicrobium thermophilum* DSM 5002 T
- Tepidamorphus geminatus* DSM 19345 T
- Lutibaculum baratangense* AMV1 T
- Methylopila capsulata* DSM 6130 T
- Rhabdaerophilum californiens* SYSU G02060 T
- Roseilacus fermentans* DSM 24875 T
- Lichenibactantia psociformis* PAMC 29148 T
- Lichenibacterium ramallinae* RmIP001 T
- Rhodoblastus acidophilus* DSM 137 T
- Methylosinus trichosporium* OB36 T
- Methylocystis parva* OBBP T
- Methylovirgula ligni* DSM 19998 T
- Methyloferula stielata* AR4 T
- Methylocapsa acidiphila* B2 T
- Bellerickia indica* ATCC 9039 T
- Pseudochelatococcus lubricantis* DSM 103870 T
- Chelatococcus asaccharovorans* DSM 6462 T
- Carnellimonas lactis* DSM 22958 T
- Salinispirillum fedricksonii* HL-109 T
- Salinarmonas rosea* DSM 21201 T
- Microvirga subterranea* DSM 14364 T
- Methylorubrum extingens* strain TK 0001 T
- Methylobacterium organophilum* NBRC 15689 T
- Enterovirga rhinocerotis* DSM 25903 T
- Bosea thiooxidans* DSM 9633 T
- Alsobacter metallidurans* CGMCC 1.12214 T
- Phreatobacter oligotrophus* DSM 25521 T
- Blastochloris viridis* ATCC 19567 T
- Xanthobacter autotrophicus* DSM 432 T
- Azorthobacter caulimodans* ORS 571 T
- Aquabacter spiritemis* DSM 9035 T
- Ancyllobacter novellis* ATCC 8093 T
- Ancyllobacter tetradsialis* DSM 5895 T
- Ancyllobacter aquaticus* DSM 101 T
- Varibacter gobiawakensis* GJM-30 T
- Rhodoplanes roseus* DSM 5909 T
- Pseudorhodoplanes sinusperisci* RPI 110 T
- Unifactor mobilis* GY H T
- Pseudobutyris taiwanensis* CCUG 51779 T
- Nitroreductor vinogradskyi* Nb-255 T
- Tardiphaga robiniae* LMG 26467 T
- Bradyrhizobium japonicum* USDA 6 T
- Afrisa carboxidovorans* OM5 T
- Alfipa felis* ATCC 53690 T
- Colhaeseibacter gelatinilyticus* DSM 18289 T
- Maritalea myrionectae* DSM 19524 T
- Cucunibacter marinus* DSM 18995 T
- Pelagibacterium natoliensis* B2 T
- Paradevosia shaoquanensis* JS-3 T
- Devosia riboflavina* JFO 13584 T
- Ascentitalea aurentiae* 42-50 T
- Salaquinomonas pukyongii* KR3-28 T
- Aureimonas altamontensis* DSM 21988 T
- Consoletella salifrons* USBA 369 T
- Mangrovicella endophytica* 5T4P-12-1 T
- Fujimarinia pelagi* HTCC2506 T
- Aurantimonas corallicola* DSM 14790 T
- Phyllobacterium myrsinacearum* DSM 5892 T
- "Paramesorhizobium deserti"* A-3-E T
- Pseudochromobacterium asaccharolyticum* DSM 25619 T
- Paenochromobacter gallinarum* DSM 22336 T
- Falsochromobacterium ovis* DSM 26720 T
- Brucella anthropi* ATCC 49188 T
- Brucella melitensis* 16M T
- Bartonella bacilliformis* KC583 T
- Zhengella mangrovei* X9-2-2 T
- Notocornibacter marinus* XM1TR244 T
- Limoniobacter endophyticus* KC7C 42097 T
- Chelatoviorans multithrophicus* DSM 9103 T
- Aquibium oceanicum* MCCC 1K02305 T
- Tianweiliana sediminis* 28 T
- Corticobacterium populi* KC7C 42249 T
- Pseudaminobacter salicylatoxidans* DSM 6986 T
- Mesorhizobium loti* DSM 2626 T
- "Oryzicola mucosus"* ROOL2 T
- Aquamicrobium delvilii* DSM 11603 T
- Aninobacter aminovorans* DSM 7048 T
- Orcolia cellulolytica* JCM 19534 T
- Roseitalea porphyridii* MA7-20 T
- Oceaniradius stylonematis* STC1 T
- Ahrensia kielensis* DSM 5890 T
- "Allopararhizobium mangrovei"* BGH/RG 6574 T
- "Neopararhizobium haloflavum"* XC0140 T
- Georhizobium profundum* WS11 T
- Pseudohoeftia suadecae* YC6898 T
- Hoeftia marina* DSM 16791 T
- Martelia mediterranea* DSM 17316 T
- Onobrychidicola muellerharveyae* TH2 T
- Shinella granulosa* DSM 18401 T
- Mycoplana citrophila* DSM 7138 T
- Xavennessaea oryzae* CGMCC 1.7048 T
- Pararhizobium glandinii* H152 T
- Sinorhizobium fredii* USDA 205 T
- Eristier adhaerens* ATCC 33212 T
- Rhizobium leguminosarum* USDA 2370 T
- Galleriella hungarica* DSM 29853 T
- Ciceribacter lividus* DSM 25528 T
- Peteryungia ipomoeae* smln9-1 T
- Allorhizobium undicola* ATCC 700741 T
- Pseudorhizobium marinum* MGJ06 T
- Neorhizobium galegae* HAABI 540 T
- Endobacterium cereale* RZME27 T
- Agrobacterium tumefaciens* B6 T
- Faunimonas pinastri* AS2C2 T
- Affilia marina* DSM 2698 T
- Prosthecomicrobium pneumaticum* DSM 16268 T
- Kaistia adipata* DSM 17808 T
- Baudia littoralis* ATCC 35022 T
- Scuibacillus lacustris* SA-279 T
- Hartmannibacter diazotrophicus* E19 T
- Pleiomorphomonas oryzae* DSM 16300 T
- Oharaebacter diazotrophicus* DSM 102969 T
- Mongoliimonas terrestris* MIMKB18 T
- Chthonobacter albugineus* KC7C 42450 T
- Rhodobium orientis* DSM 11290 T
- Pseudovibrio denitrificans* DSM 17465 T
- Roseibium denhamense* JCM 10543 T
- Pannonibacter phragmitetus* DSM 14782 T
- Stappia stellulata* DSM 5886 T
- Hongsoonnocoella zoyisae* SY4-7 T
- Bregmatia corrubedonensis* DSM 23382 T
- Segnochromobacter spiritosus* Sp-1 T
- Pseudoxanthobacter soli* DSM 19599 T
- Amorpha coralli* DSM 19760 T
- Acuticoccus yangtzensis* JL1095 T

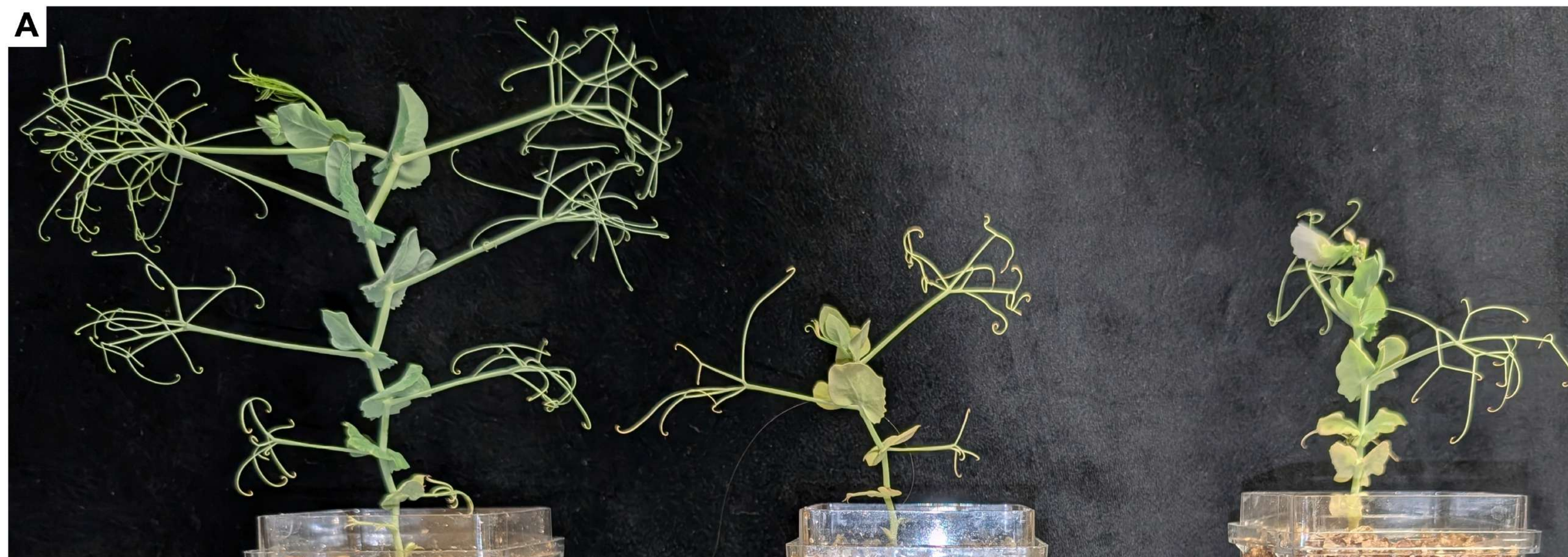
[illegible]

Alpha-rhizobia

A polyphyletic group of nitrogen-fixing symbionts of legumes, spread across six families of the order *Hyphomicrobiales*

A recent article claimed that *Sphingomonas sediminicola* DSM 18106^T (order *Sphingomonadales*) is also a rhizobium

However, our data is inconsistent with this claim



R. johnstonii 3841^T

S. sediminicola DSM 18106^T

Uninoculated

Genome-based phylogeny of the bacterial order *Hyphomicrobiales*



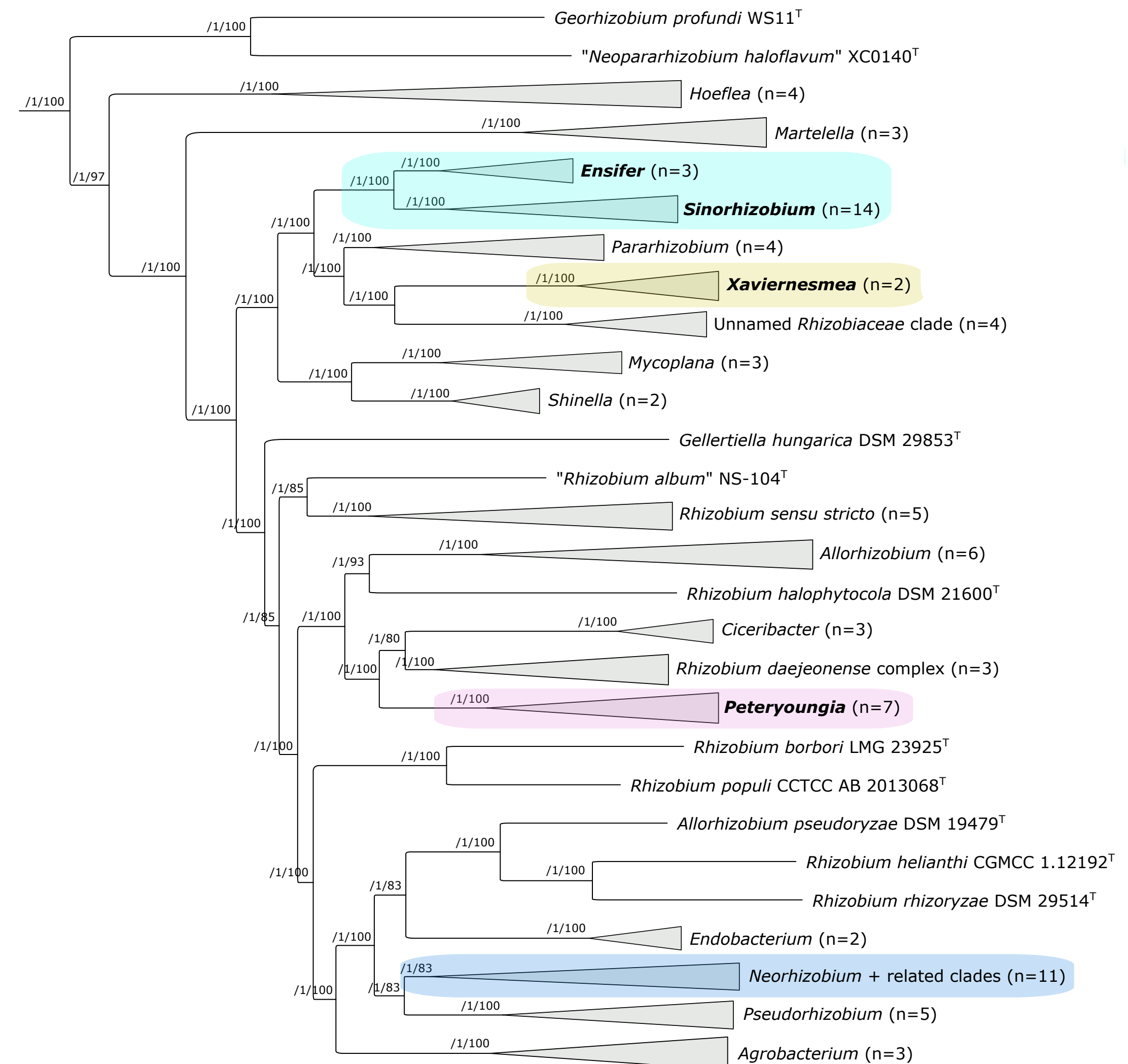
The family *Rhizobiaceae*



Now encompasses ~280 species across nearly 50 genera

Includes many plant symbiotic bacteria (rhizobia) and pathogens (agrobacteria)

Historically, this family contained many taxonomic inconsistencies

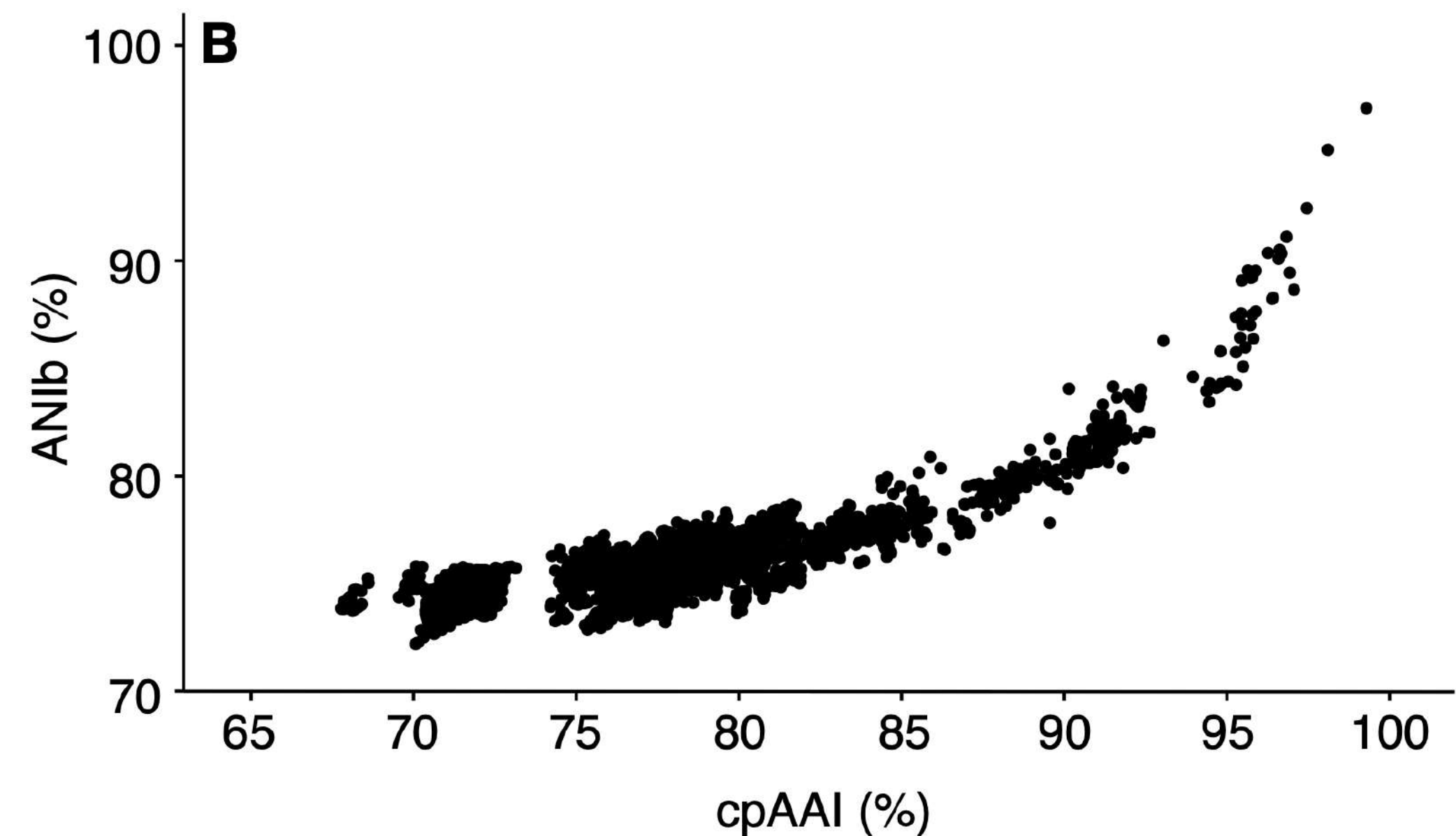
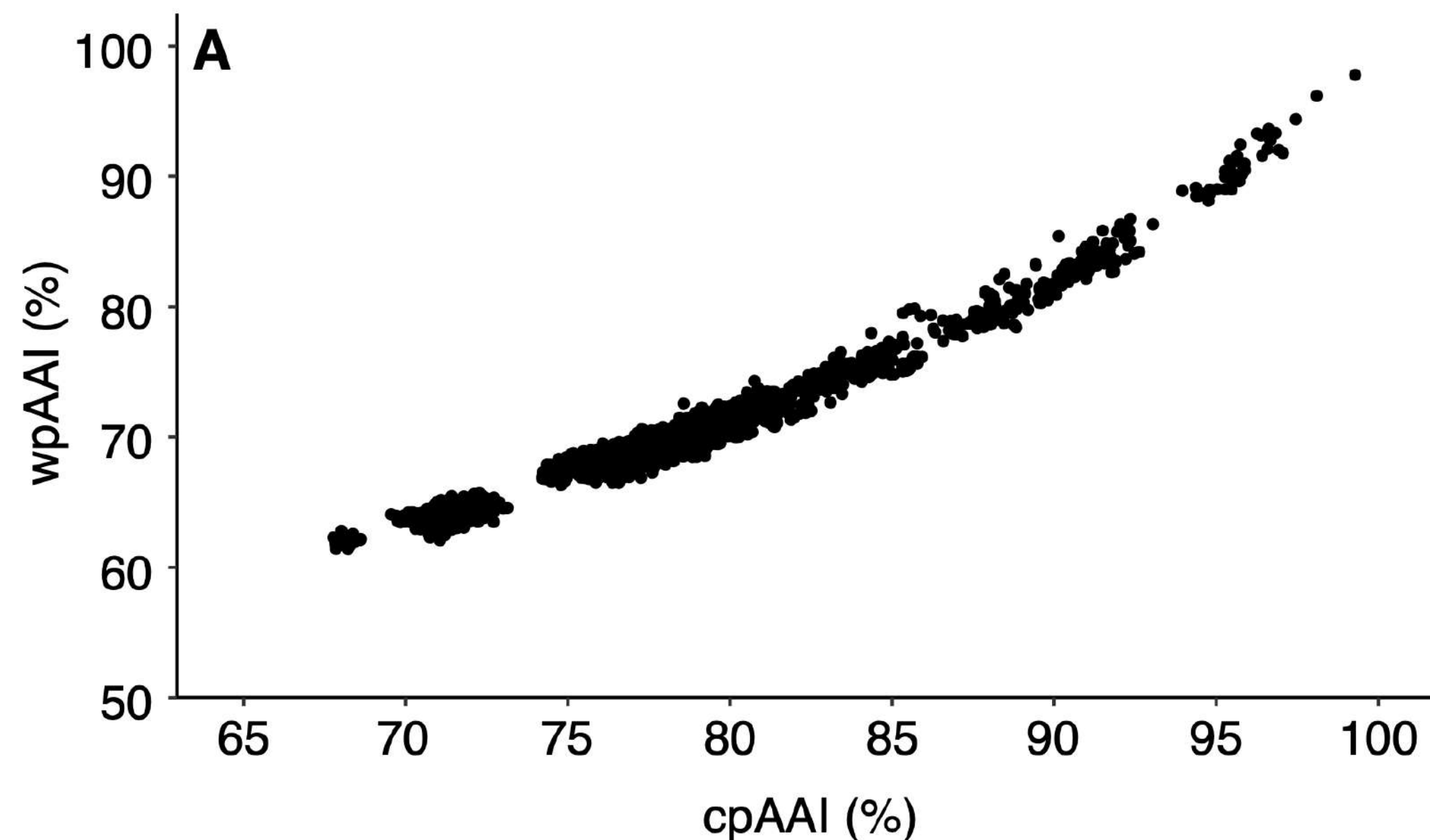


Defining genus boundaries in the family *Rhizobiaceae*



Identified a core set of 170 single-copy, non-recombining genes across 94 representative type strains

Used for phylogenomics and to identify a core-proteome AAI of ~86.0% (~86.5%) as a proposed, flexible threshold for genus demarcation



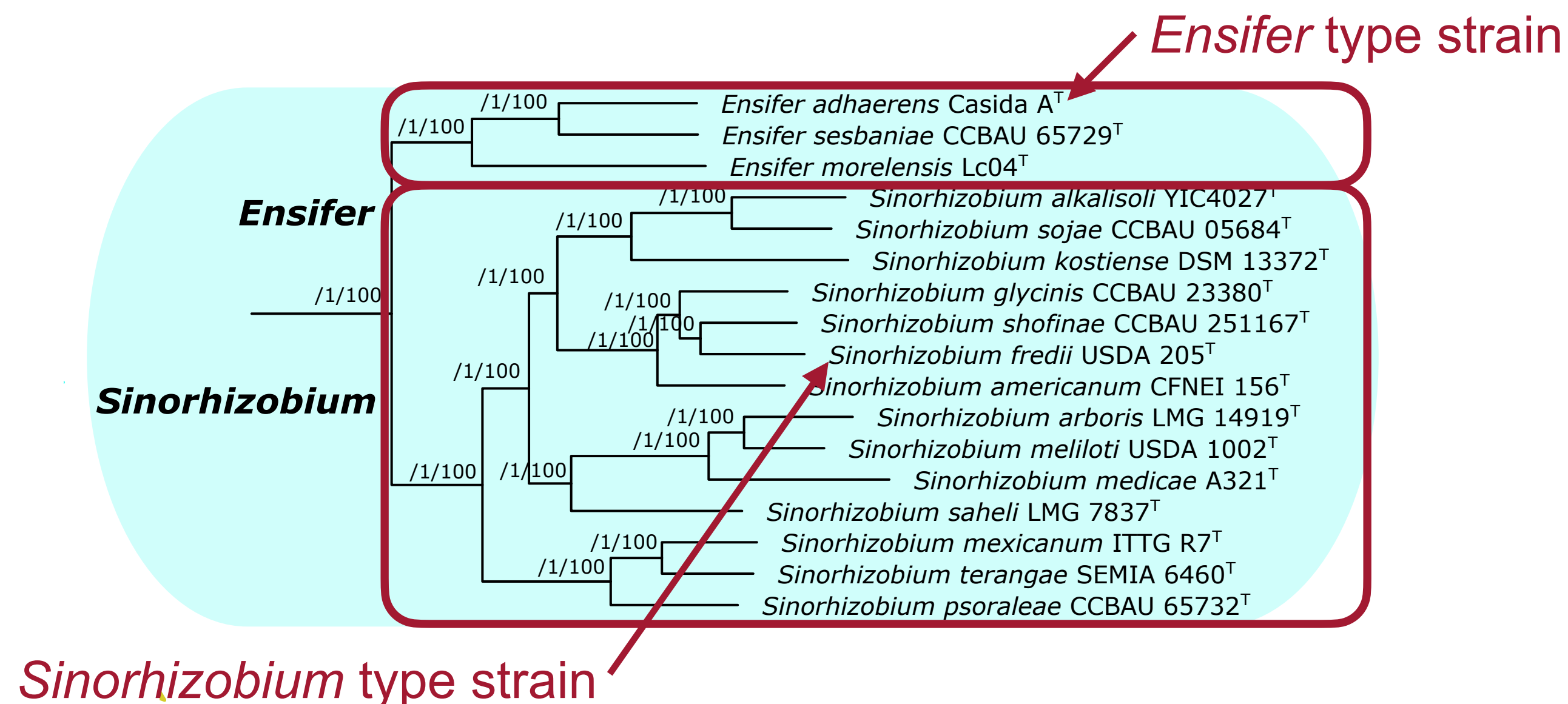
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Led to proposal for one new genus, several new combinations, and to argue that *Sinorhizobium* and *Ensifer* are distinct genera



Defining genus boundaries in the family *Rhizobiaceae*



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Led to proposal for one new genus, several new combinations, and to argue that *Sinorhizobium* and *Ensifer* are distinct genera

But many species missing from this initial analysis

Further revisions to the taxonomy of the family *Rhizobiaceae*

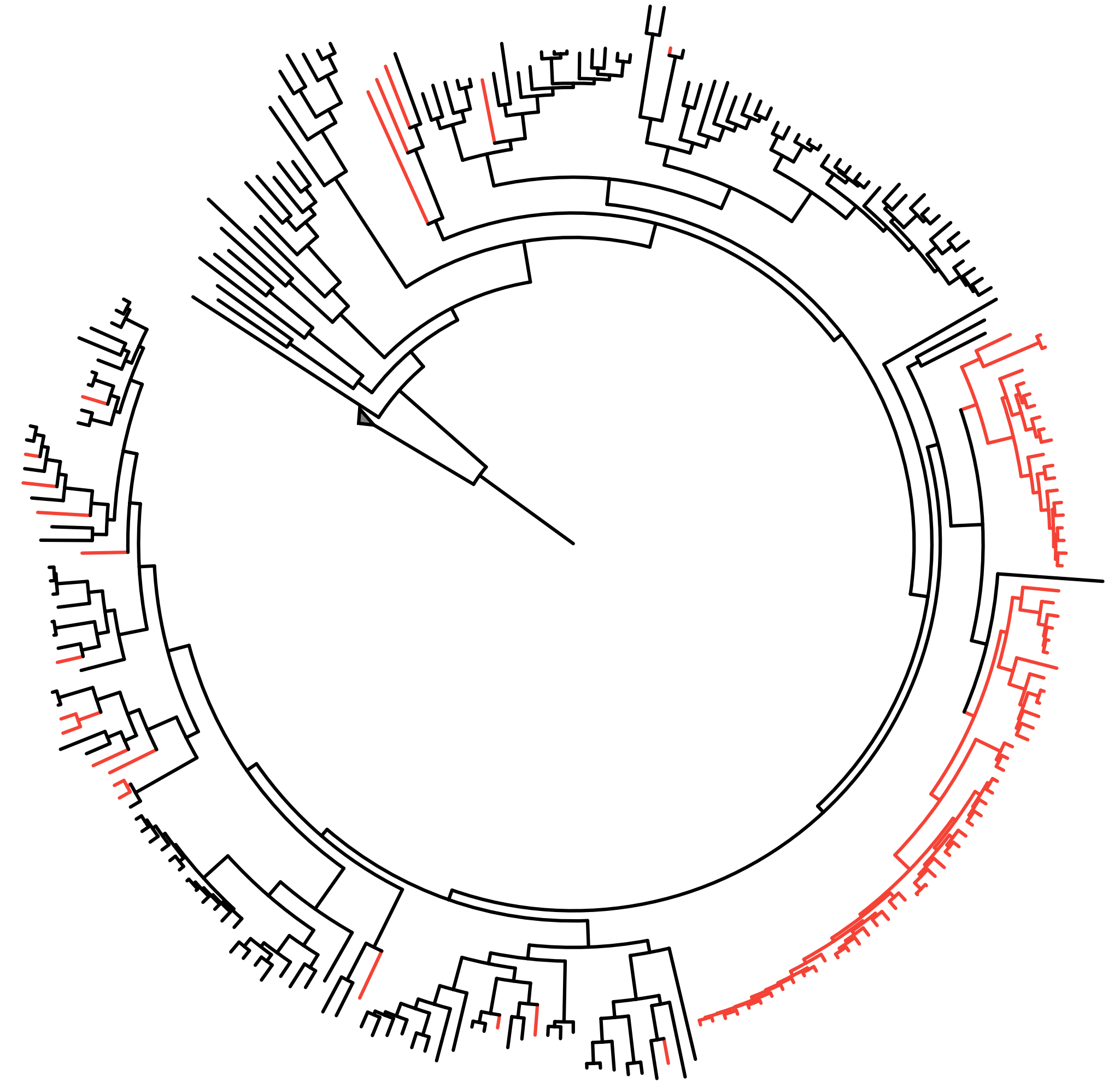


Performed whole genome sequencing of 14 *Rhizobiaceae* type strains lacking publicly available genome sequences

Analyzed 242 *Rhizobiaceae* type strains (up from 94)

Dozens of *Rhizobium* species were clearly distinct from the genus *Rhizobium* and thus required reclassification

Proposed 8 novel genera and 32 novel combinations



Phylogeny of 242 *Rhizobiaceae* species type strains

Further revisions to the taxonomy of the family *Rhizobiaceae*

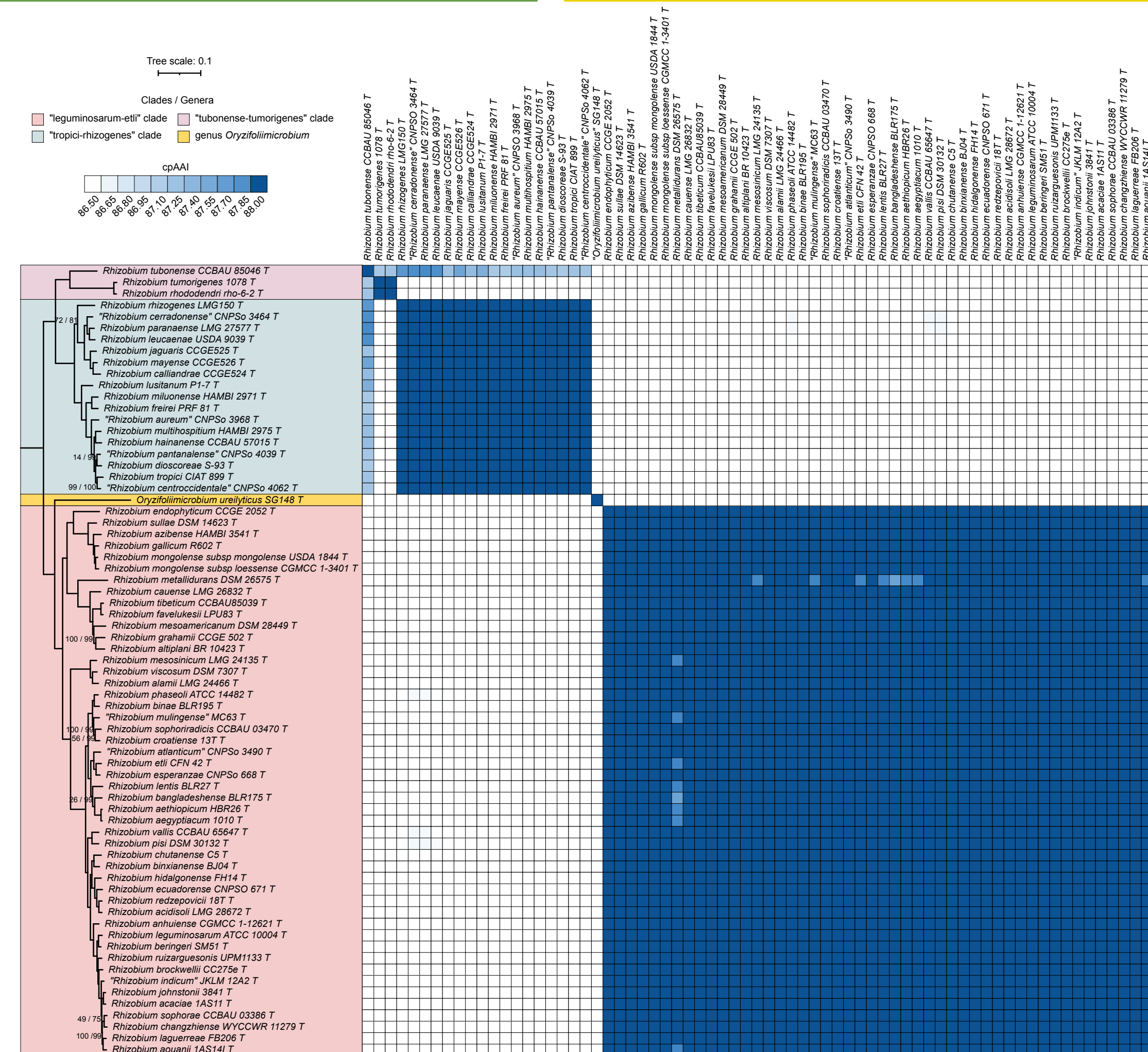


Rhizobium can be split into three clades

Most within-clade cpAAI comparisons are $> 88\%$ while most between-clade cpAAI comparisons are $< 86.5\%$

Rhizobium is paraphyletic due to a recently proposed genus, *Oryzifoliimicrobium*

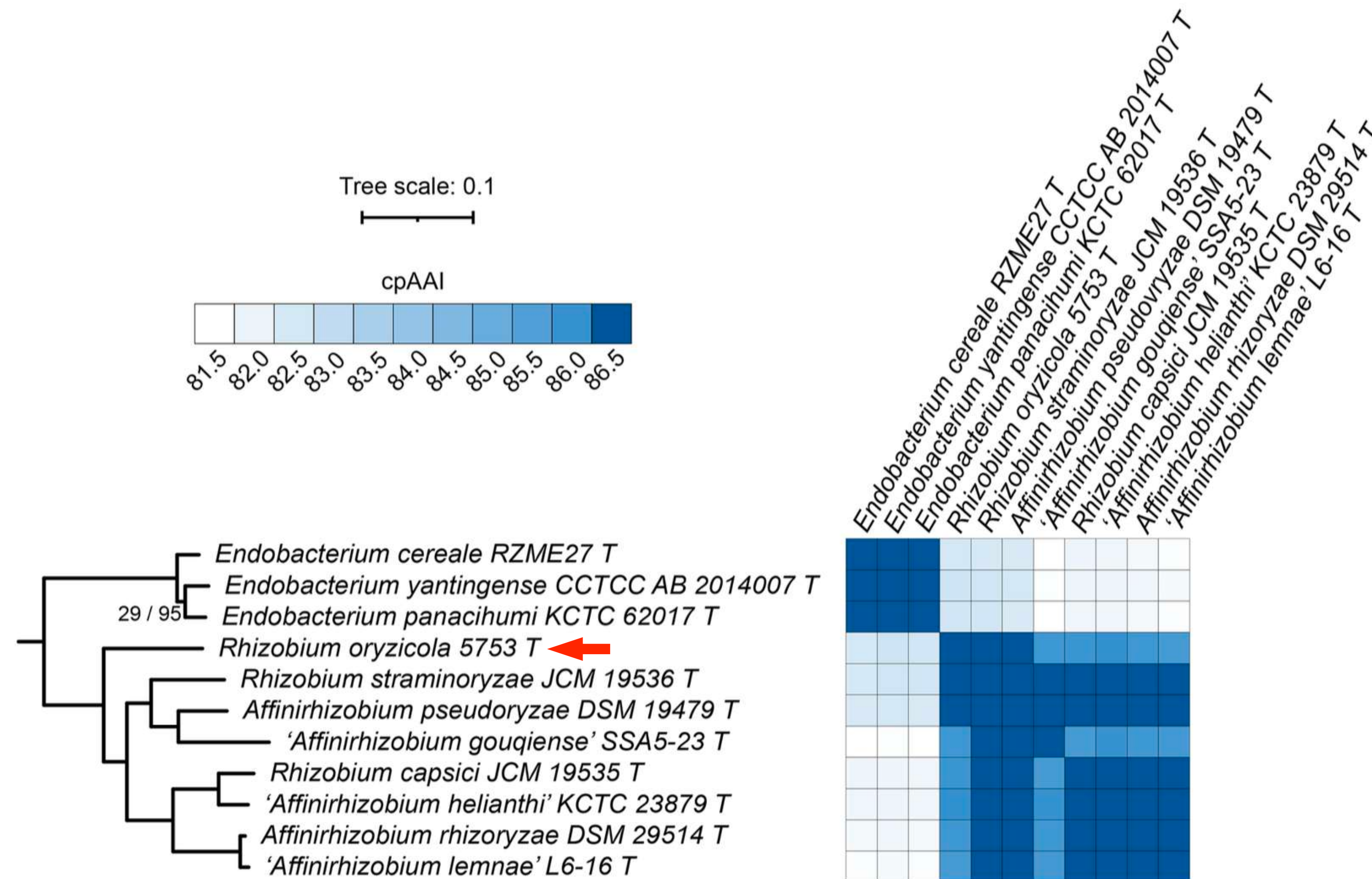
Thus, the three core *Rhizobium* clades are proposed to represent distinct genera (*Rhizobium*, *Martinezella* gen. nov., and *Ariminella* gen. nov.)



Phylogeny and cpAAI of *Rhizobium* and related genera

Next steps in *Rhizobiaceae* taxonomy

Sequence additional *Rhizobiaceae* type strains and check taxonomic assignments
Use SeqCode to validate novel combinations where needed



The people who actually do the research



Current Lab Members

Dr. Tia Harrison (TMU)

Dr. Sabhjeet Kaur

Dr. Upkar Pandher

Oona Esme

Kristy Moniz

Ameneh Nourozian

John Robinson

Mia Rondinelli

Gaia Rota

Nicholas Smith

Jesse Branje

Ariane Mayrand Nicol

Natalia Naranjo

Erika Gagnon

Erin Griffiths

Emma Klimczak

Abby Duggan

Dev Patel

Adam New



The BENEFIT Team



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Dr. George diCenzo



Dr. Terry Bell



Dr. Matthew Bakker



Dr. Derek Brewin



Dr. Warren Mabey



Dr. Neal Scott



Dr. Rebecca Doyle



Dr. Ana Vargas



Dr. Jill Hobbs



Dr. Yvonne Lawley



Dr. Olivia Wilkins



Dr. Nicholas Corradi



Dr. Marina Cvetkovska



Dr. Allyson MacLean



Dr. Sabine Liebenehm



Dr. Yang Yang

Culture collections (in no particular order)



DSMZ (Germany) and Dr. Katharina Huber-Fischer

LMG (Belgium)

JCM (Japan)

KCTC (South Korea)

NBRC (Japan)

CCUG (Sweden)

HAMBI (Finland)

USDA (USA)

Funders and Partners



Government



GenomeCanada



GenomePrairie



Ontario Genomics

Manitoba



Ontario



Industry



fieldless

Grower Organizations



MANITOBA
CROP
ALLIANCE



Research Institutions



University
of Manitoba



Queen's
UNIVERSITY



UNIVERSITY OF
TORONTO
SCARBOROUGH



uOttawa



UNIVERSITY OF
SASKATCHEWAN



Other

