



# How to define a prokaryotic species

Joint Graduate Seminar

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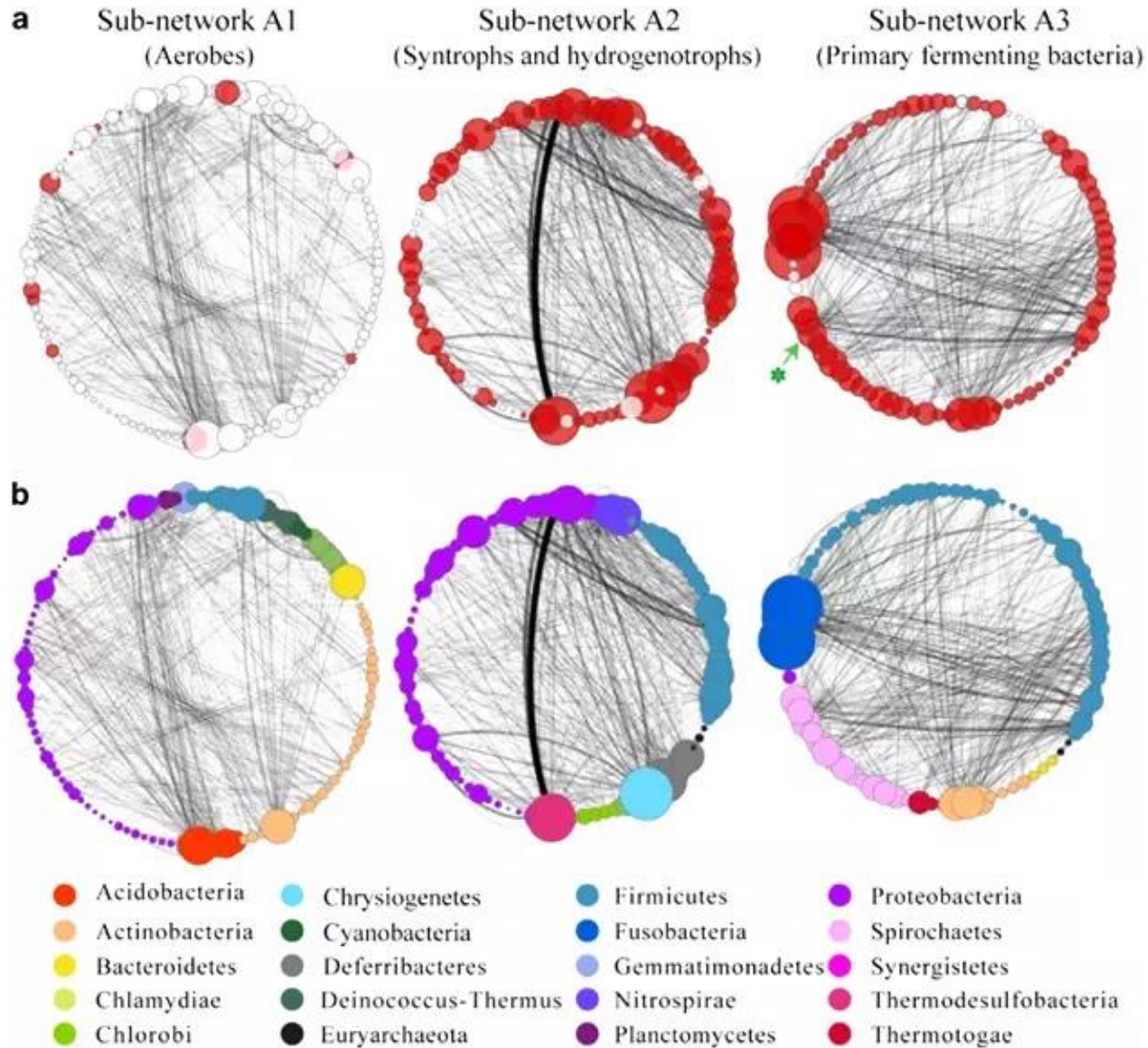
**Department:** Microbiology

# Outline



- 01** Challenges of applying the classical definition to prokaryotes
- 02** Practical importance and operational definitions
- 03** Concepts considering speciation mechanisms
- 04** Defining species and populations under the scope of microbial speciation

# Challenges of applying the classical definition to prokaryotes



- Genetic exchange network among distantly related taxa
- Edges: the number of transferred genes
- Nodes: the genomes from different phyla

# Practical importance and operational definitions

## Necessity:

- Clinical settings
- Agriculture
- Food industry
- ...

## 16S rRNA gene

Cutoff: 97%[1]

DNA-DNA  
Hybridization  
(DDH)  
experiments

Since 1960s, the gold standard:

## DNA-DNA binding

Cutoff: 70% [1]

## Average Nucleotide Identity (ANI)

Cutoff: 95% [1]

Ribosomal  
sequencing

## Polyphasic approach:

Combination of genotypic (DNA fingerprinting), phenotypic (biochemical tests, fatty-acid composition) properties and phylogenetic information (rRNA gene sequences)

A set of **single copy conserved genes** [2]

Whole genome  
sequencing

Shotgun  
metagenomics

[1] Goris et al. Int J Syst Evol Microb. 2007

[2] Truong et al. Nat. Methods. 2015

# Practical importance and operational definitions






nature  
biotechnology

RESOURCE

<https://doi.org/10.1038/s41587-020-0501-8>



## A complete domain-to-species taxonomy for Bacteria and Archaea

Donovan H. Parks  , Maria Chuvochina, Pierre-Alain Chaumeil, Christian Rinke , Aaron J. Mussig  and Philip Hugenholtz 

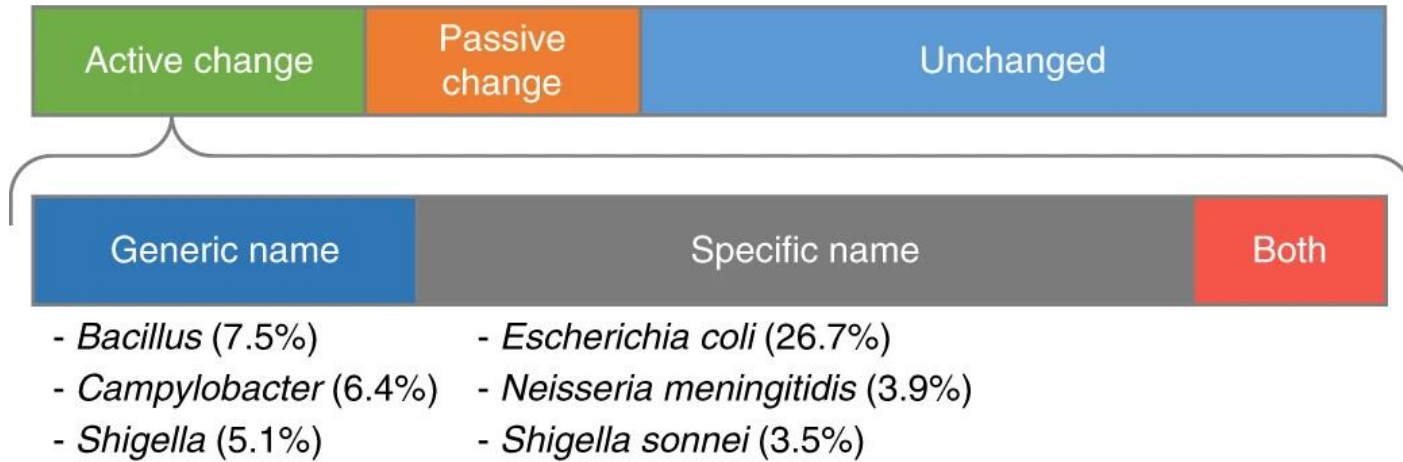
The Genome Taxonomy Database is a phylogenetically consistent, genome-based taxonomy that provides rank-normalized classifications for ~150,000 bacterial and archaeal genomes from domain to genus. However, almost 40% of the genomes in the Genome Taxonomy Database lack a species name. We address this limitation by using commonly accepted average nucleotide identity criteria to set bounds on species and propose species clusters that encompass all publicly available bacterial and archaeal genomes. Unlike previous average nucleotide identity studies, we chose a single representative genome to serve as the effective nomenclatural 'type' defining each species. Of the 24,706 proposed species clusters, 8,792 are based on published names. We assigned placeholder names to the remaining 15,914 species clusters to provide names to the growing number of genomes from uncultivated species. This resource provides a complete domain-to-species taxonomic framework for bacterial and archaeal genomes, which will facilitate research on uncultivated species and improve communication of scientific results.

- Pairwise ANI comparison against representative genomes
- Greedy clustering for de novo species clusters

Parks et al. Nat. Biotechnol. 2020

# Results of the nomenclature for the unassigned genomes

**a**



Unchanged: the genome's binomial species name was identical in the NCBI taxonomy

Passively change: the NCBI taxonomy did not have a species assignment

Active change:

- (1) Generic name: change in only the generic name of the species
- (2) Specific name: change in only the specific name of the species
- (3) Both: changes in both the generic and specific names

**b**

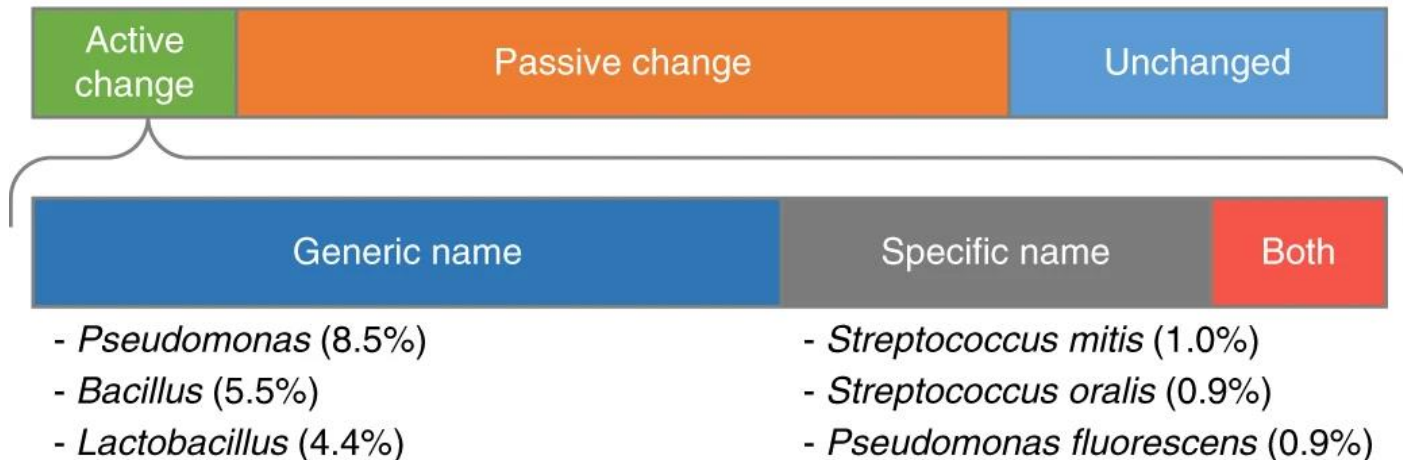


Fig. Results are shown for the 143,566 genomes (a) and 24,080 species representatives (b) with an NCBI taxonomic assignment.

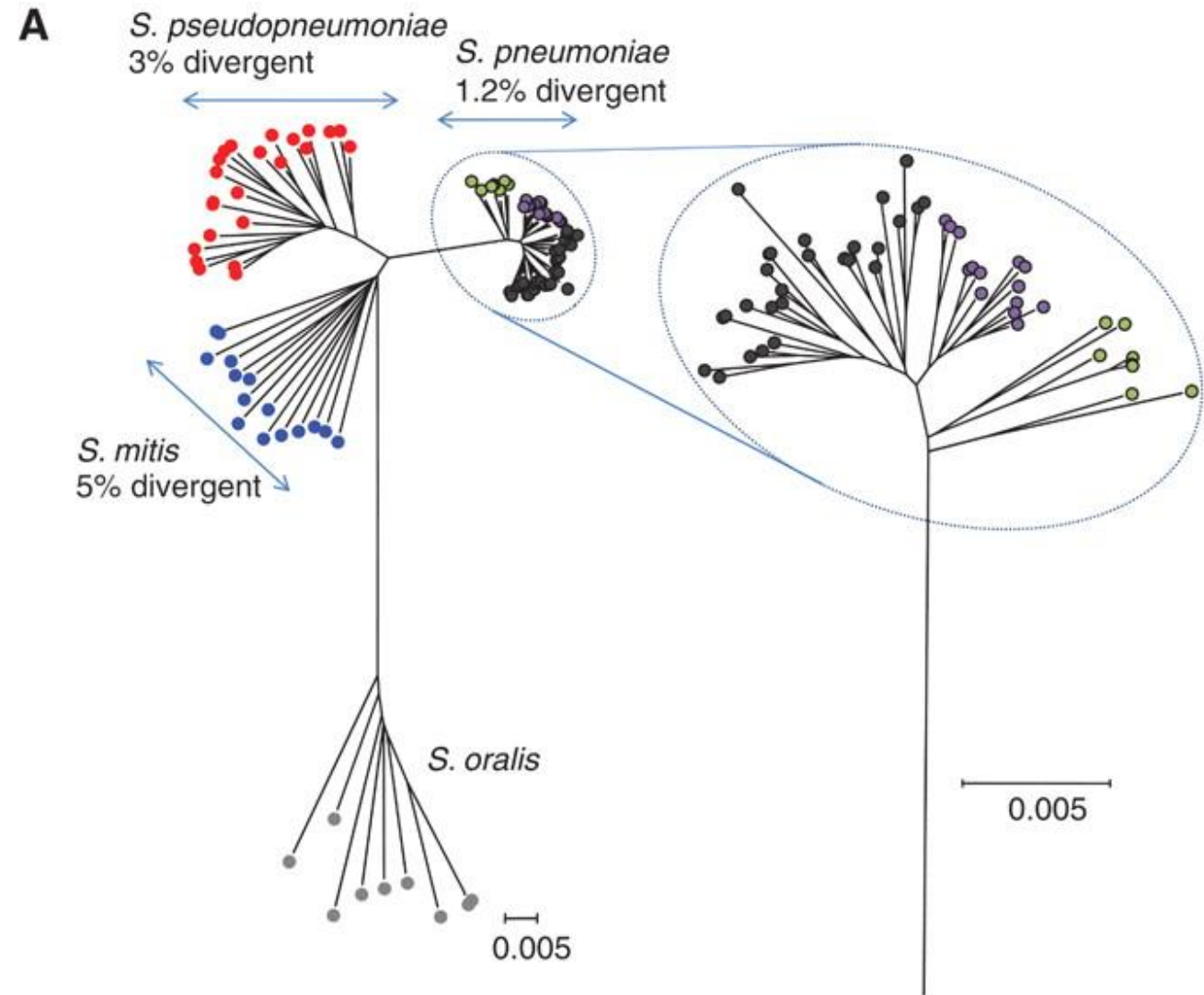
# Problems of the operational definitions

Fig. The evolution tree of 97 isolates of four Streptococcus species. The tree was built using concatenates of six housekeeping loci.

*S. pneumoniae*: a major human pathogen

*S. mitis*: a commensal bacteria

*S. pseudopneumoniae*: a recently described organism of uncertain status



# Concepts considering speciation mechanisms

- The Ecological Species Concept (ESC): speciation is driven by **natural selection**
- The Biological Species Concept (BSC): speciation is driven by barriers to genetic exchange, which means rates of **gene flow (recombination)** are high within than between species, but cross-species gene transfer can still occur.

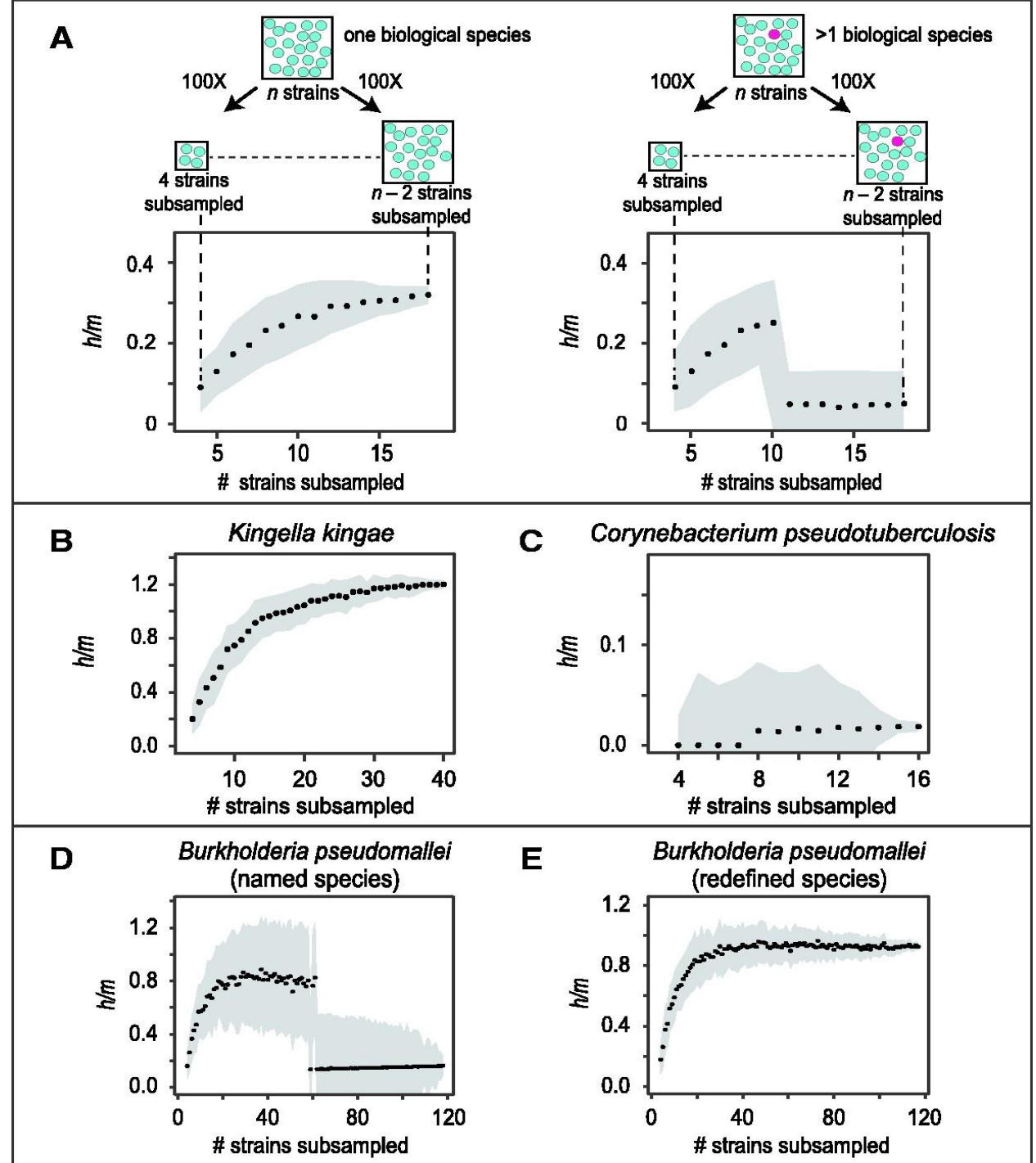


# Redefining prokaryotic species by quantifying the gene flow (BSC-like)

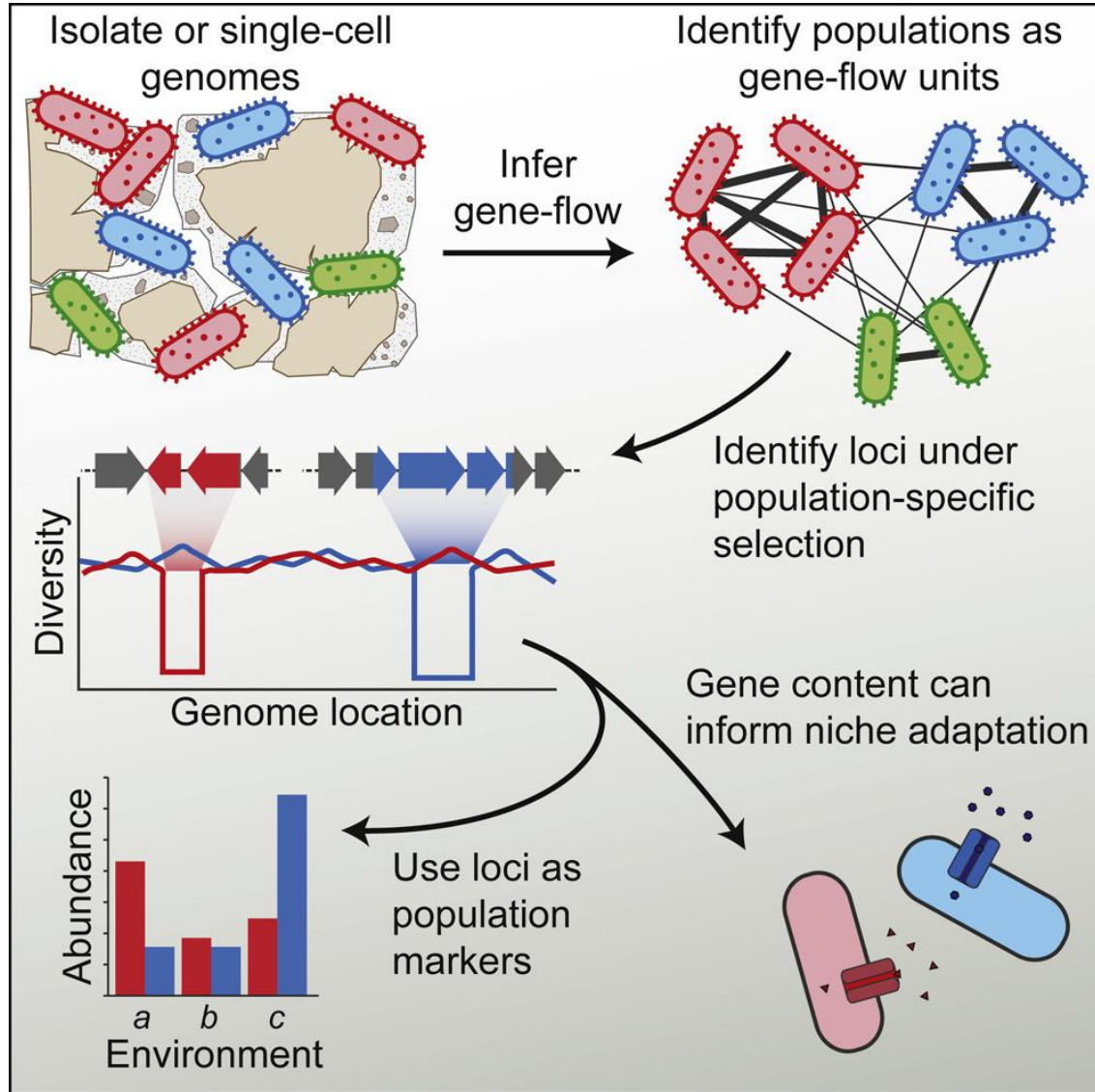
$h/m$ : the ratio of homoplasious to nonhomoplasious alleles

## Rationale:

- (1) If a group of strains are from one biological species, the gene flow will be coherent.
- (2) If there are any strains from different species, a break of gene flow will occur.



# Defining bacterial populations considering the speciation mechanisms

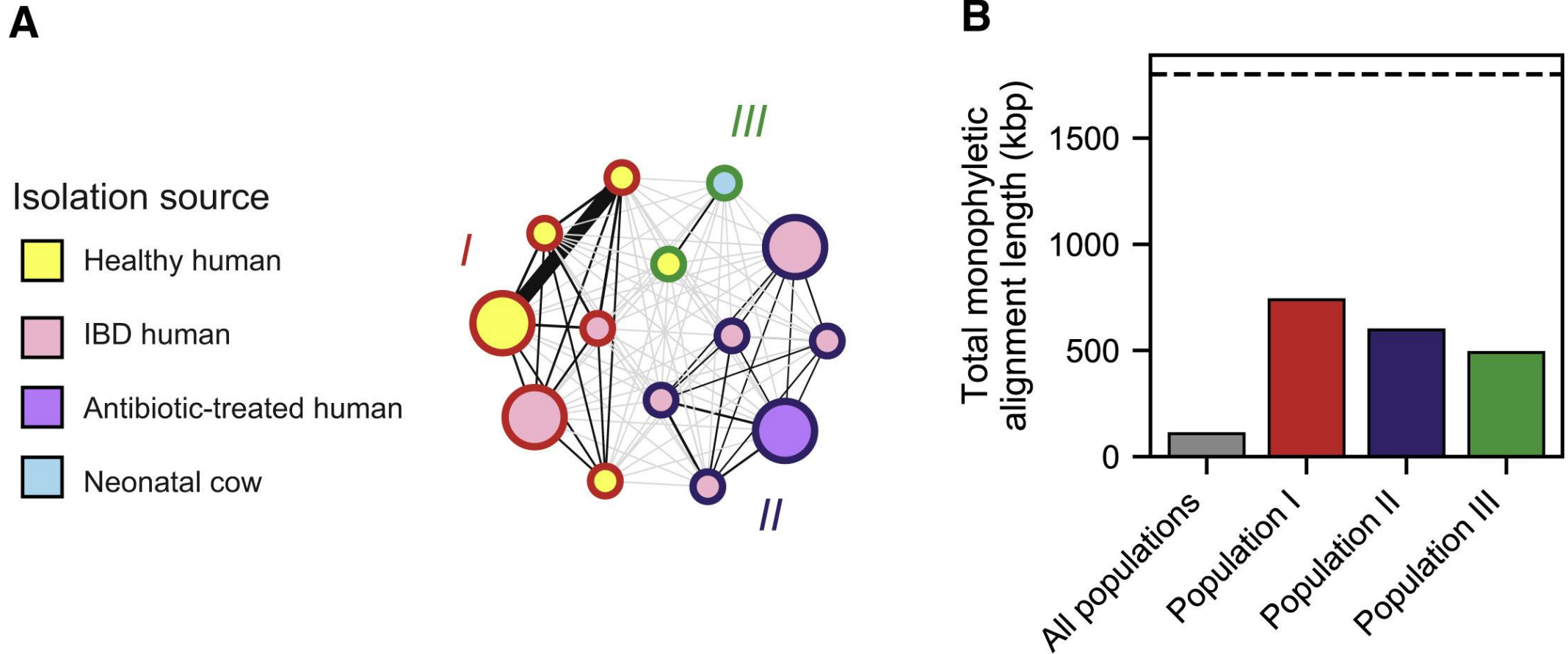


Species: gene flow units

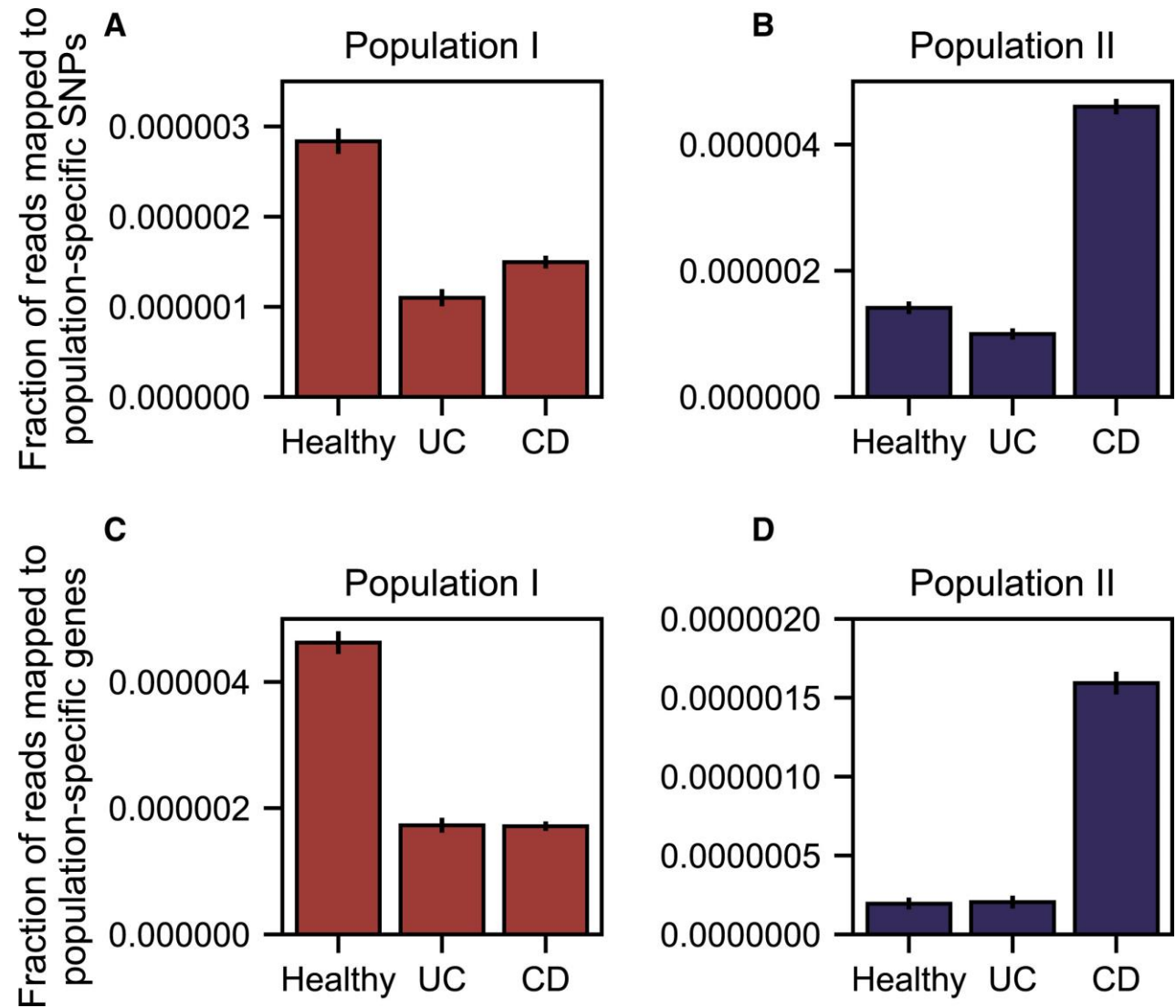
Populations: **locally co-existing** members of a species that are reproductively isolated

Recent research suggested that distinct populations can be identified based on cohesive ecological dynamics ([natural selection](#)) and preferential gene flow ([recombination](#)).

# Fine Scale Population Structure Is Evident in *R. gnavus* Populations



Verification of the *R. gnavus* Populations with metagenomic reads coverage



# Concluding remarks

- Ordering microbial diversity into ecologically and genetically cohesive units is both theoretically and practically necessary
- Without considering the speciation mechanisms, scientists demarcate the prokaryotes into species according to the genomic similarities.
- Defining prokaryotic species and populations considering speciation mechanisms