Traditional Nepali Fermented Foods as Reservoirs of Functionally Relevant Microbes

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"Unlocking functional microbes from traditional fermentation using genomics and phenotyping"

Background

- Traditional Nepali fermented foods are widely consumed and culturally embedded
- Potential sources of:
  - Vitamin-producing microbes
  - Gut-beneficial bacteria
- Functional traits remain **poorly characterized**

Objective

- To identify genome-encoded nutritional and gut-relevant traits in bacteria from Nepali fermented foods.

Methodology (Overview)

Study Design and Dataset Overview

- 363 isolates from 35 traditional fermented foods
- 231 genomes sequenced
- 210 high-quality genomes retained

96 non-redundant representative strains (99.99% ANI)

Workflow:
Sampling → Isolation → Whole-genome sequencing → Annotation → Functional screening

Methodology (Functional Screening)

Genomic screening (in silico):

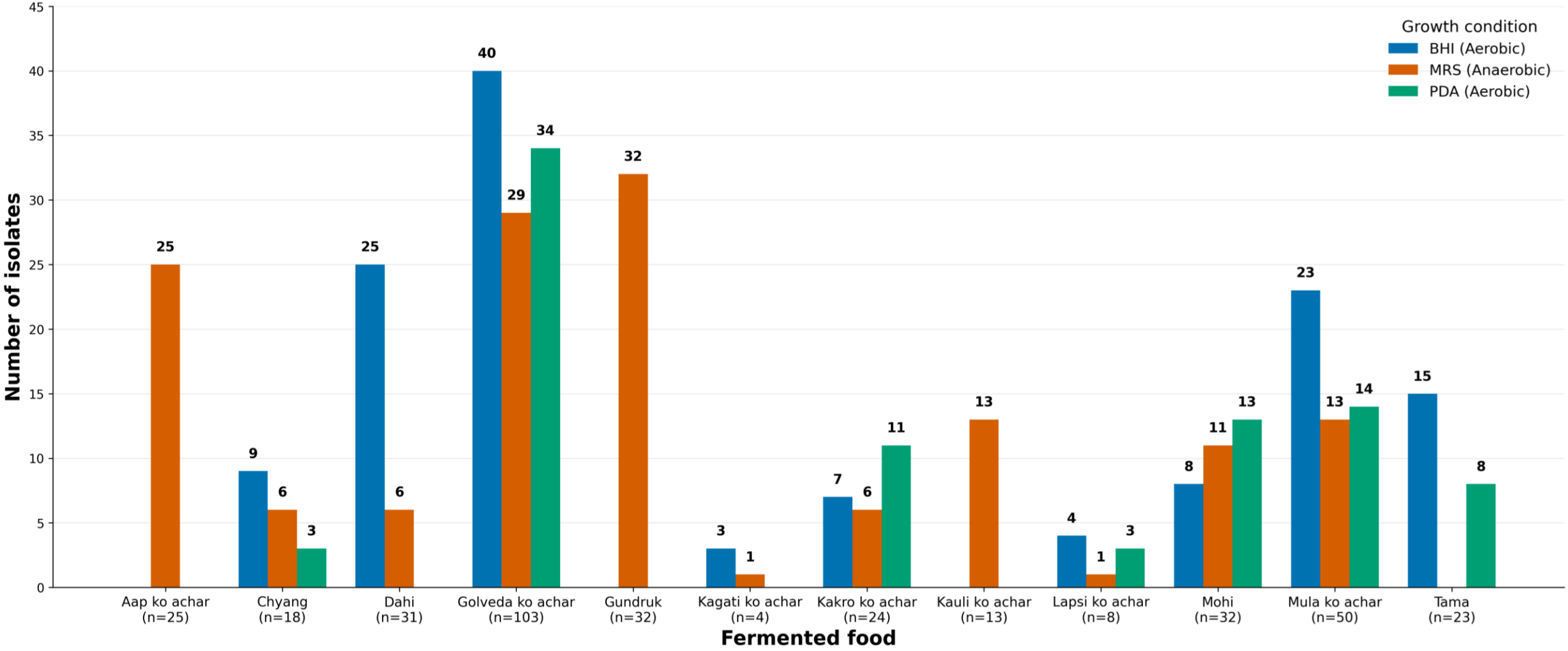
- GABA biosynthesis pathways
- Vitamin biosynthesis (B1, B2, B7, B9, B12)
- Bacteriocin related genes
- EPS and biosurfactant-related genes
- Stress tolerance genes

Phenotypic validation (in vitro):

- Acid, bile tolerance and bile salt hydrolase (BSH) activity
- Antimicrobial activity (MRSA, *E. coli*)

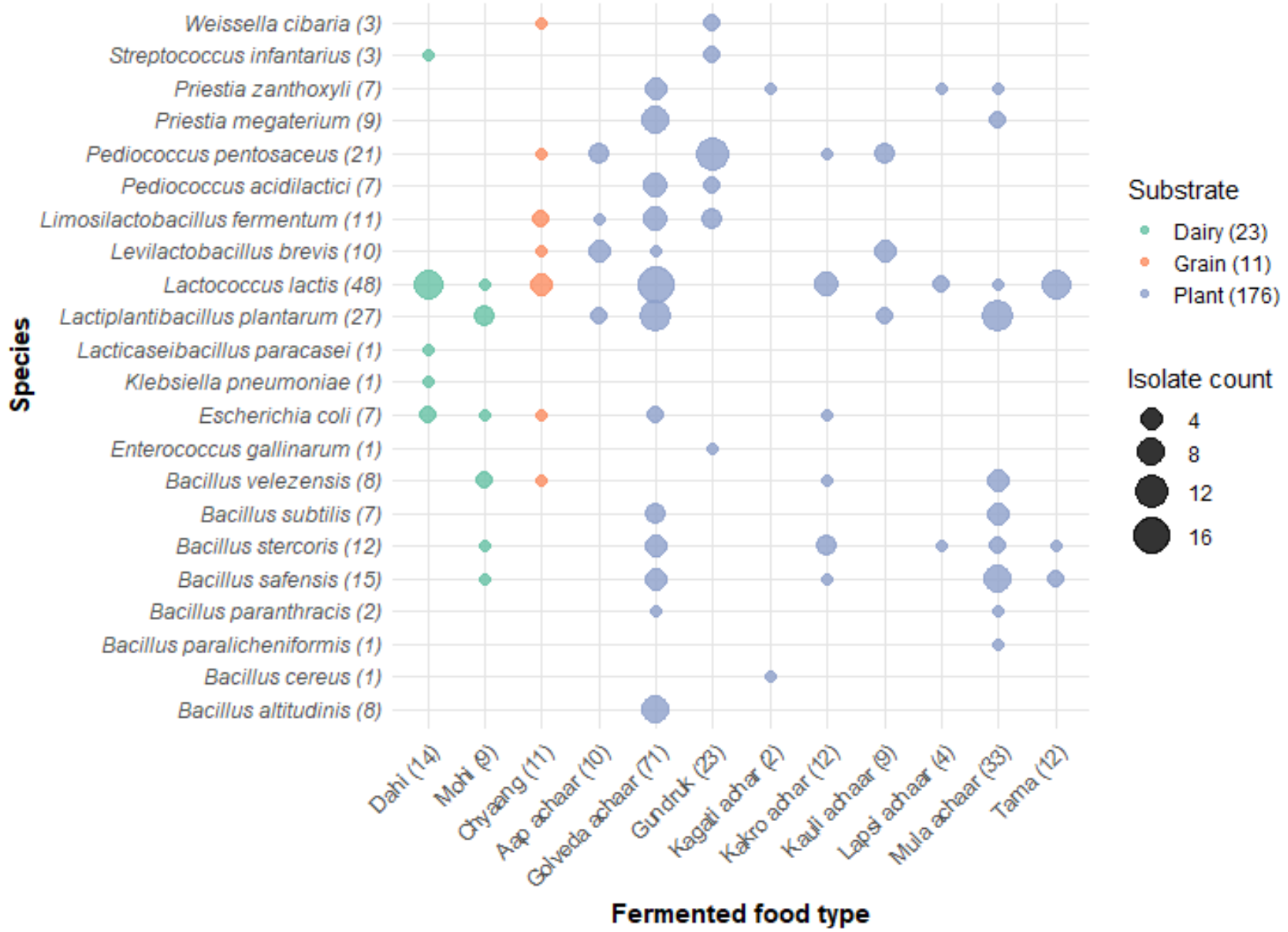
Integrated genomic and phenotypic analyses were used to identify functionally promising strains

Microbial Isolation Across Nepali Fermented Foods



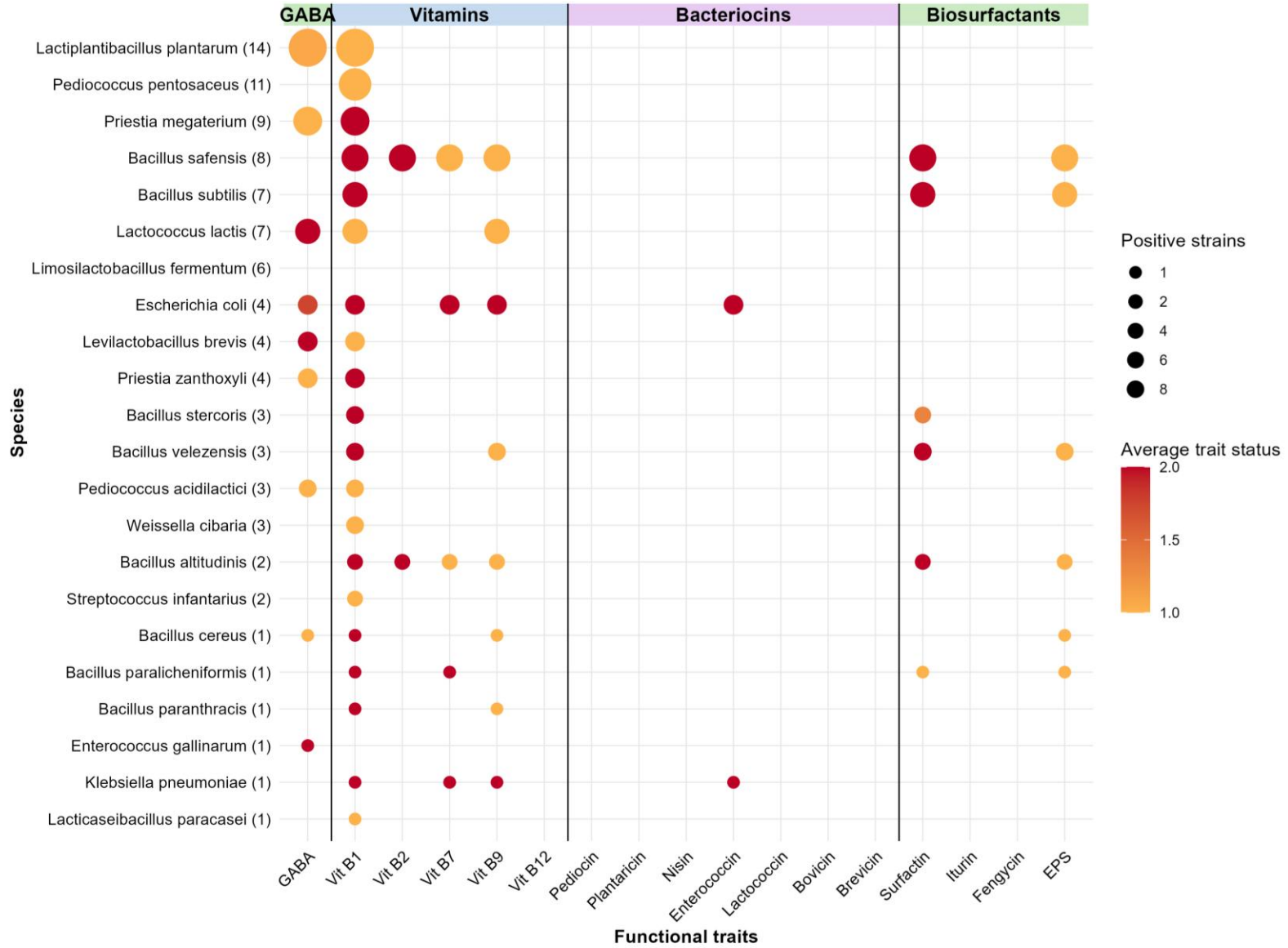
Achar samples showed the highest isolate diversity

Taxonomic Diversity Across Fermented Foods



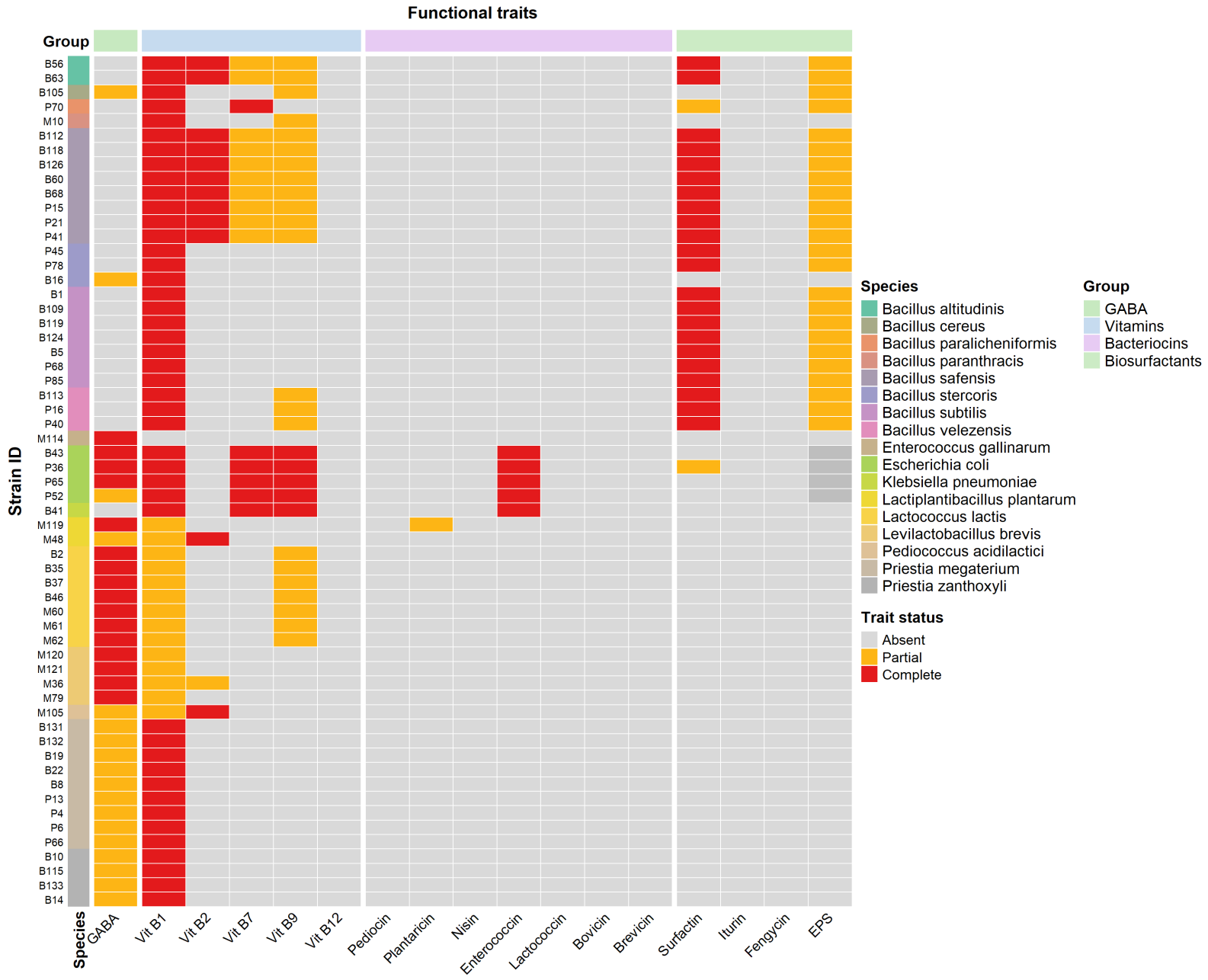
Plant-based fermentations show highest diversity, dominated by LAB and Bacillus

Species-level Functional Potential of Fermented Food Isolates



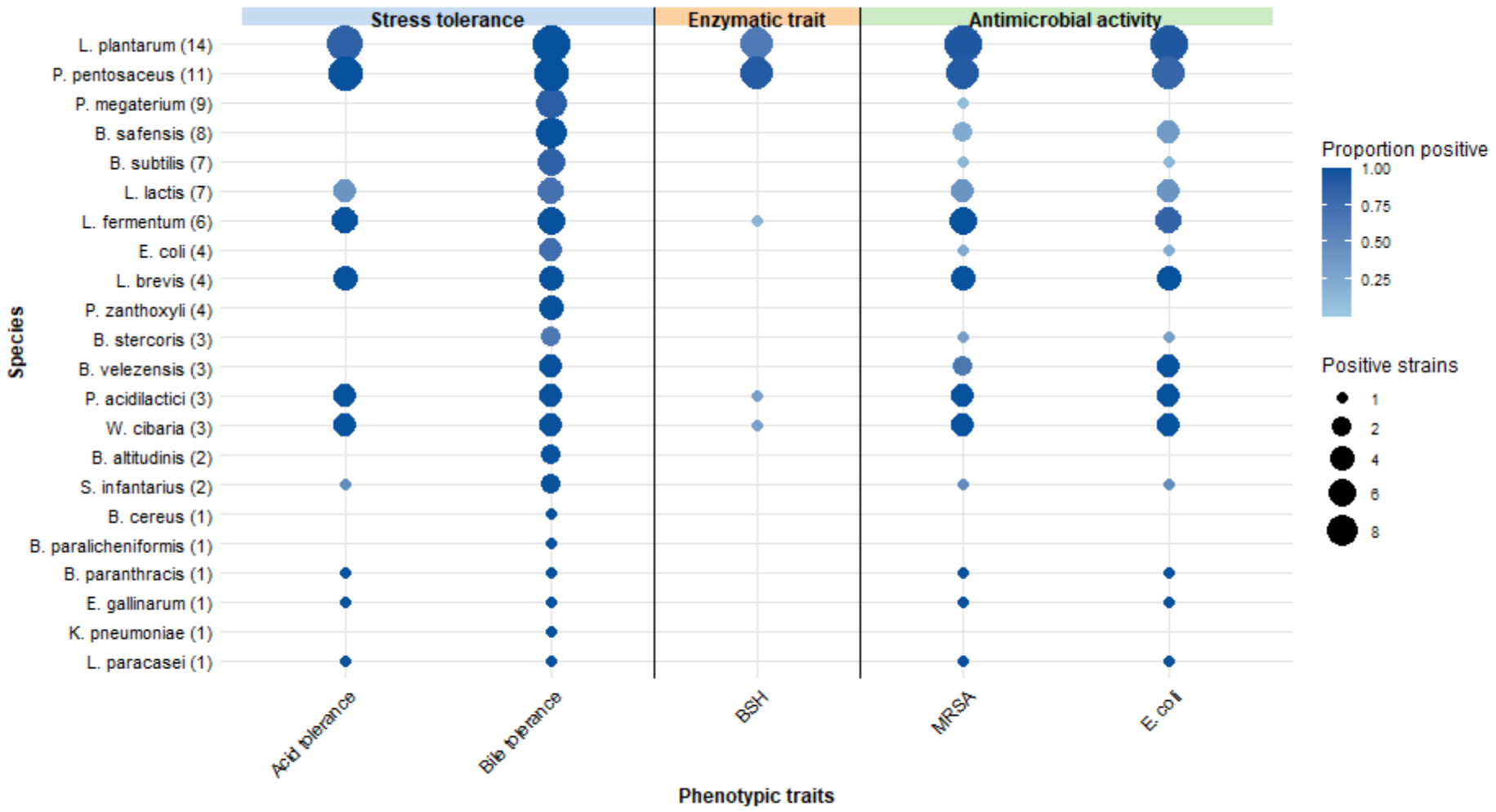
Complete pathways were unevenly distributed across taxa, with **Bacillus spp. showing the strongest functional completeness**, while **LAB contributed selectively, particularly for GABA-related traits.**

Strain-Level Distribution of Functional Traits



Functional potential varies markedly across strains, with uneven distribution of complete and partial pathways across trait categories.

Species-Level Distribution of Phenotypic Traits



Stress tolerance was more broadly distributed across taxa, whereas BSH and antimicrobial activity were restricted to subsets of species.

Strain-Level Distribution of Functional Traits Across Isolates

Phenotypic traits varied across strains and showed significant associations with functional genes, including *bsh* and stress-response genes (*katA*, *sodA*), with antimicrobial activity showing moderate genomic correlations, highlighting strain-level functional diversity.

Conclusion: Takeaway Messages

- Nepali fermented foods harbor functionally rich microbial diversity
- Identified strains with vitamin production and gut survival potential.
- Representative strains include *L. fermentum* & *L. brevis* (GABA), *L. plantarum* (gut survival), *Pediococcus/Lactococcus* (antimicrobial), *Bacillus* spp. (vitamins)
- Supports development of low-cost functional foods
- Strong relevance for nutrition security in Nepal.

Acknowledgment

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Presenter

Shrijana Shakya is a Research Scientist at Sabitri Sciences Pvt. Ltd., Nepal, with a PhD in Probiotic Sciences from Hiroshima University, Japan. Her research focuses on exploring functional microbes from traditional fermented foods using integrated genomic and phenotypic approaches, with the aim of developing low-cost, locally relevant probiotic and functional food solutions. She is actively involved in advancing microbiome research in Nepal through the KiKha1K (Himalayan Fermented Food Microbiome) Project.