

Southeast Dairy Producer's Check-Off Program Research Summary

Development of customized effective probiotics to mitigate dairy calf diarrhea

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Implications

Neonatal calf diarrhea (NCD) is a gastrointestinal disorder causing significant mortality and economic loss in calves under one month old, with about 57% of weaning calf deaths attributed to it. Despite improvements in the dairy industry, NCD remains a persistent problem due to its multifactorial nature, driven by infections from various enteric pathogens. Our study aims to find a treatment for NCD in dairy calves using probiotics from healthy newborn calves. We analyzed fecal samples from both healthy and diarrheic calves and found that Bifidobacteria were significantly higher in healthy calves and negatively associated with septicemia. These Bifidobacteria strains, unique to calves, present a promising treatment opportunity. Animal trials will assess the efficacy of these probiotics in treating NCD and septicemia, offering an alternative to antibiotics. Successful implementation could reduce calf diarrhea and the associated economic losses, including calf mortality, treatment costs, and impaired growth.

Methods

A total of 130 fecal samples, including 47 from healthy calves and 82 from diarrhea calves, were collected from the rectal-anal junction of dairy calves using an anaerobic collection method. 16S rRNA sequencing was performed to determine correlation between bacterial abundance in fecal samples and health outcomes of dairy calves. Bifidobacteria strains were isolated from healthy dairy calves using BSM agar and identified by sangar sequencing. Strains identified as Bifidobacteria were conducted for whole-genome sequencing and assembled using Sickle for trimming, and SPAdes genome assembly through the UFRC Galaxy pipeline. A core-genome alignment was generated by Roary; a core-genome based phylogenetic tree was generated using IQ-TREE, and the generated tree was visualized by iTOL. The virulence factors were identified through the BV-BRC and the antimicrobial resistance genes were determined through the Comprehensive Antibiotic Resistance Database. Genes associated with stress response were pinpointed using data from published research and by annotation of the genome with Prokka.

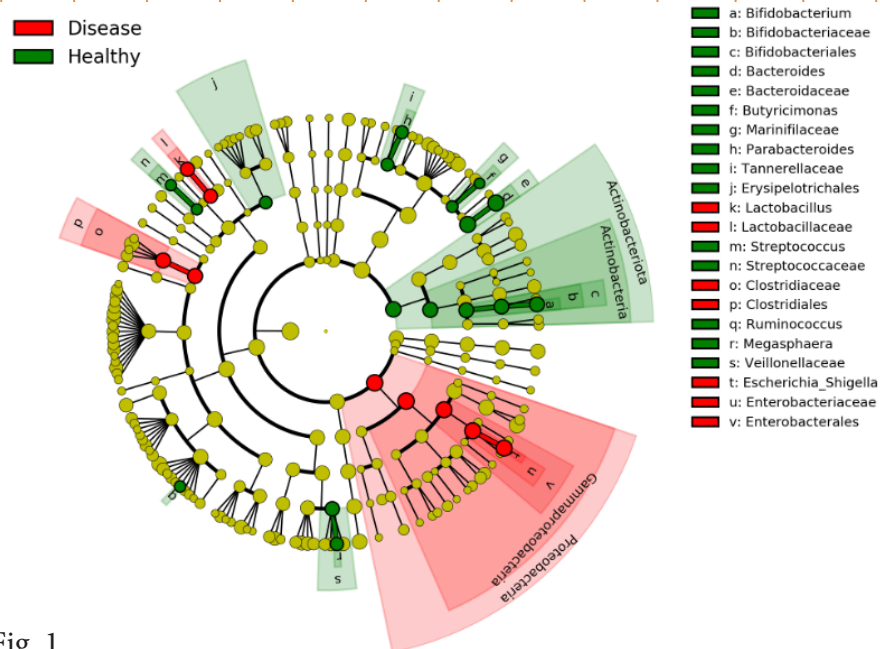


Fig. 1.

Results

Through 16S rRNA gene sequencing analysis, we found a strong negative correlation between the abundance of Bifidobacteria and septicemia incidence in calves, suggesting that specific Bifidobacterium strains could be effective probiotics to prevent sepsis in diarrhea calves. From healthy calves, we isolated 54 *B. longum* and 6 *B. pseudocatenulatum* strains. By comparing our *B. longum* genomes to 50 other publicly available *B. longum* genomes, we found host- and environment-related genomic features. Using in silico analysis, we tested whether these strains carry antimicrobial resistance genes (ARGs). All tested strains carried ARGs, suggesting these strains might have acquired them from antibiotic use on dairy farms. After conducting in vitro tests, selected probiotic candidates will be used to treat calves with diarrhea and septicemia.

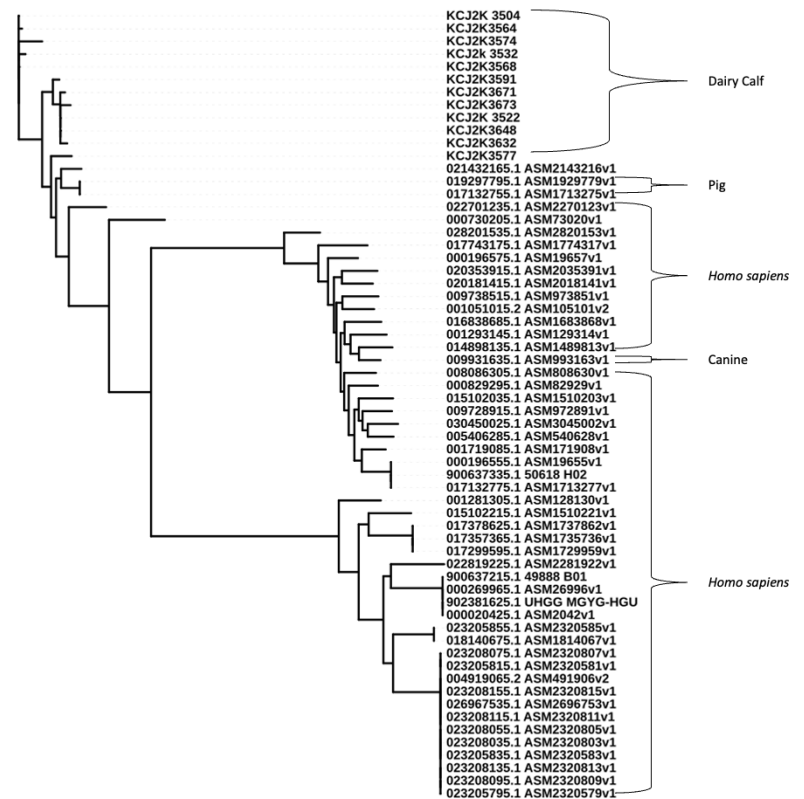


Fig. 2. Phylogenetic tree of Bifidobacterium isolated from dairy calves.

References of Published Work

Crabill, H., Y. Zhai, b. Choi, F. Peixin, and K.C. Jeong. Identification of Bifidobacteria with probiotic potential for the treatment of septicemia in dairy calves. EPI Research Day. Gainesville, FL. Feb. 2024.