



Searching for the Perfect Pick: Screening the Bacterial Isolates from Pickles for Selection the Top Strain for Microgreens Inoculation

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Introduction:

Microgreens, the young seedlings of edible plant species, boast various health benefits and temptate consumers with their unique appearance and flavorful taste. Despite their numerous health benefits, microgreens are highly vulnerable to abiotic stresses, particularly drought, leading to rapid loss of freshness and susceptibility to rot and wilting. Current literature offers limited guidance on addressing these challenges, focusing primarily on interventions like altering light spectra, modifying packaging, and fortifying with nutrients. However, research on microbial species, particularly bacteria, reveals promising mechanisms for plants to adapt to drought conditions. *Bacillus* species, commonly associated with drought-tolerant plants, have shown positive effects on crop yields under drought stress. Our study aims to select and identify strains of *Bacillus subtilis* and *Bacillus coagulans*, isolated from dairy and fermented products, to enhance the quality, shelf-life, and drought resistance of microgreens.

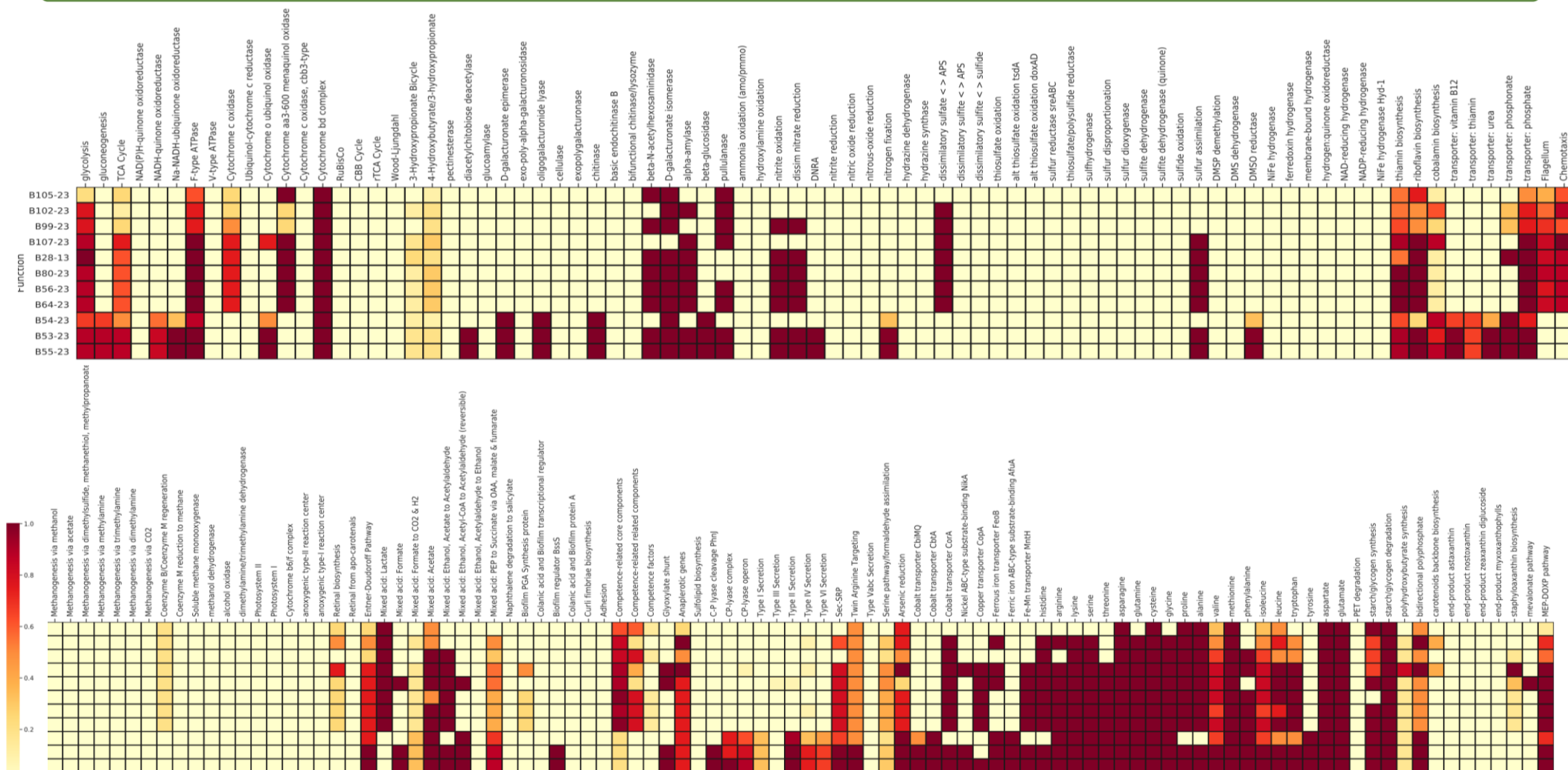
Hypothesis:

Microgreens inoculated with selected *B. subtilis* or *B. coagulans* strain will improve their parameters, such as quality, resistance and shelf-life, under the drought stress conditions.

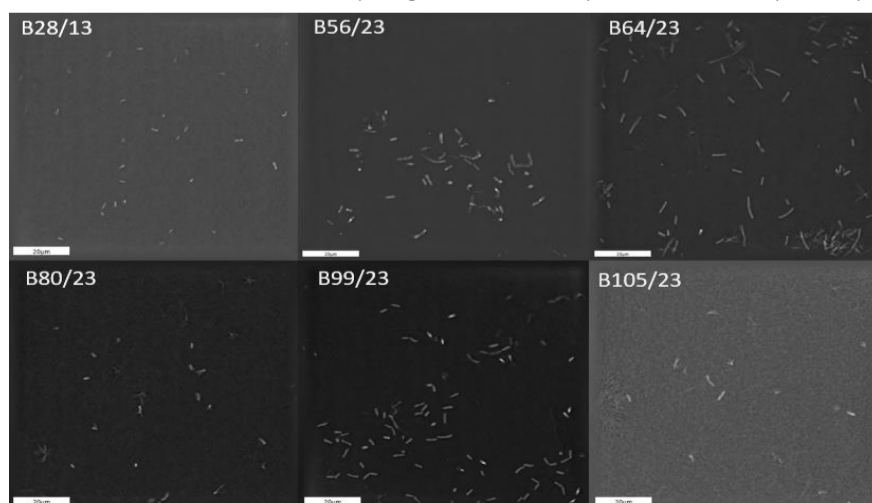
Materials and methods:

A total of 96 strains isolated from dairy and pickled products were preliminarily identified using the PCR method with two specific markers: endonuclease for *B. subtilis* detection and comK for *B. coagulans*. All strains underwent Sanger sequencing for the 16S region to confirm PCR identification. Based on the obtained sequences, phylogenetic trees were created using MEGA Software. From these results, 11 strains confirmed as *B. coagulans* or *B. subtilis* were selected for whole genome sequencing using Illumina next-generation sequencing technology. The resulting sequences were used to create two heatmaps: one for the presence of resistance gene clusters and another for active genes associated with various metabolic pathways. Based on these analyses, 6 strains were chosen for further investigation. This included whole transcriptome sequencing using Illumina technology, metabolic analysis using the BIOLOG GENIII plate system, and cell morphology analysis using the Nanolive Holotomograph screening method.

Preliminary results:

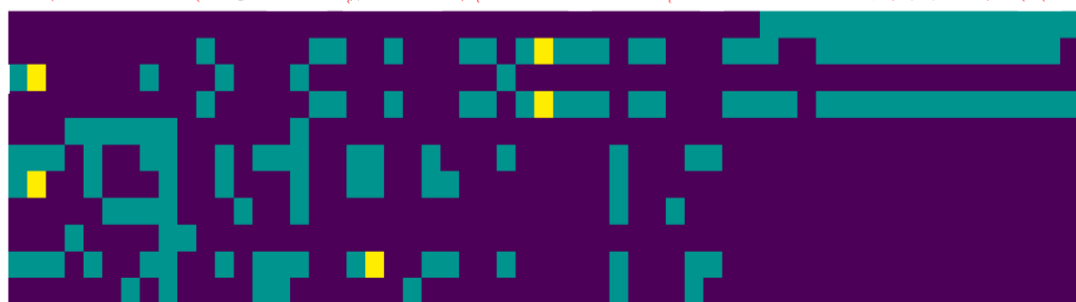


Heatmap of genes linked to specific metabolic pathways



Morphology analysis with Nanolive Holotomograph

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Heatmap of resistance genes identified in studied bacterial strains genomes