

EVALUATION OF THE NEUROPROTECTIVE ACTIVITIES IN GABA-PRODUCING POTENTIAL PROBIOTIC STRAINS FROM INDIGENOUS FERMENTED FOODS

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Introduction & Aim

- Gut-Brain Axis (GBA), a bidirectional network, is influenced by metabolites from the diet and fermentative activities of gut microbes. The GBA and behaviours are often linked to neurotransmission.
- γ -amino butyric acid (GABA), an inhibitory neurotransmitter, has a calming effect. During depression, a multitude of signals in our brains are generated, leading to an overload of neurotransmitters, which can lead to fatigue and other depressive symptoms.
- GABA from lactic acid bacteria has received extensive attention from both academia and industry in recent years because of the “GRAS” status of the lactic acid bacteria and their crucial roles in the food industry
- GABA-producing Lactobacilli are reported to reduce anxiety behaviours and markers of depression in vivo due to alterations in the expression of GABA receptor subunits regulating mood and anxiety, such as the hippocampus and amygdala.
- Commercial Anxiolytic drugs available exert adverse side effects.

Aim: To isolate the GABA-producing culture from fermented food and test its probiotic and neuroprotective properties.

Materials and Methods

Isolation of bacterial culture from fermented foods. Cultures were grown in MRS and screened for GABA production.

GABA confirmation through TLC, LCMS and NMR.

Isolated GABA-producing strain 16s rRNA identification by sequencing and BLAST.

Standard probiotic attributes were tested.

Neuroprotection study using neuronal cell line SH-Sy5y against: High glucose, glutamate, and lipopolysaccharide stress

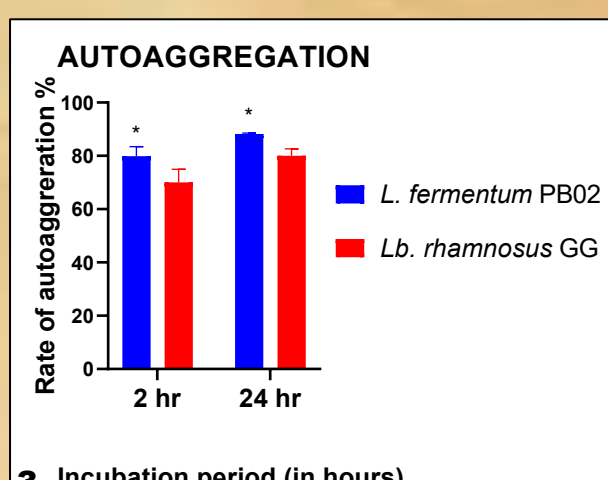
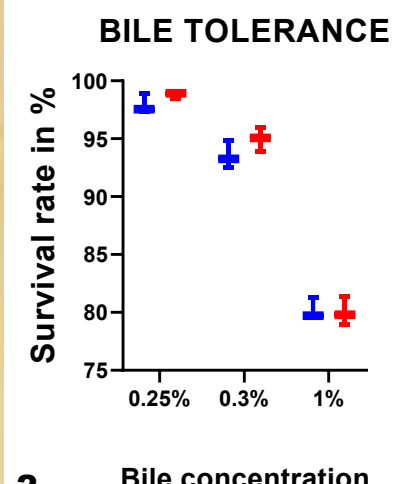
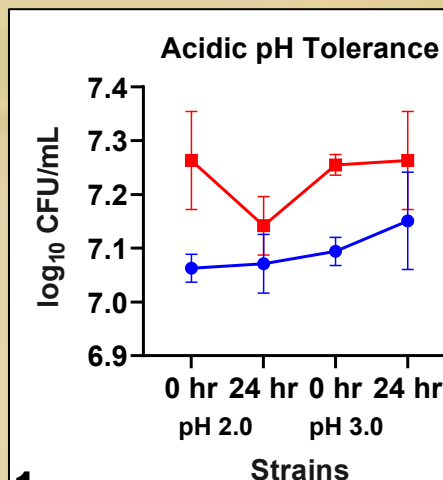
Data analysis

Future Perspectives

- GABA-producing probiotics with neuroprotective ability can be further explored for the development of GABA-enriched functional foods
- Validation of results using stress-induced in vivo models,
- Mechanisms and pathways can be further elucidated by targeting the GABAergic system.

Results

A GABA-producing culture was isolated from Fermented rice (pantha bhaat); widely consumed in the eastern part of India. The ability of the isolate to produce GABA was confirmed using TLC, LC-MS, and ¹H NMR, along with GAD assay.



Figures 1, 2, 3: Probiotic attributes of the isolate against low pH, increasing bile, and auto-aggregation.

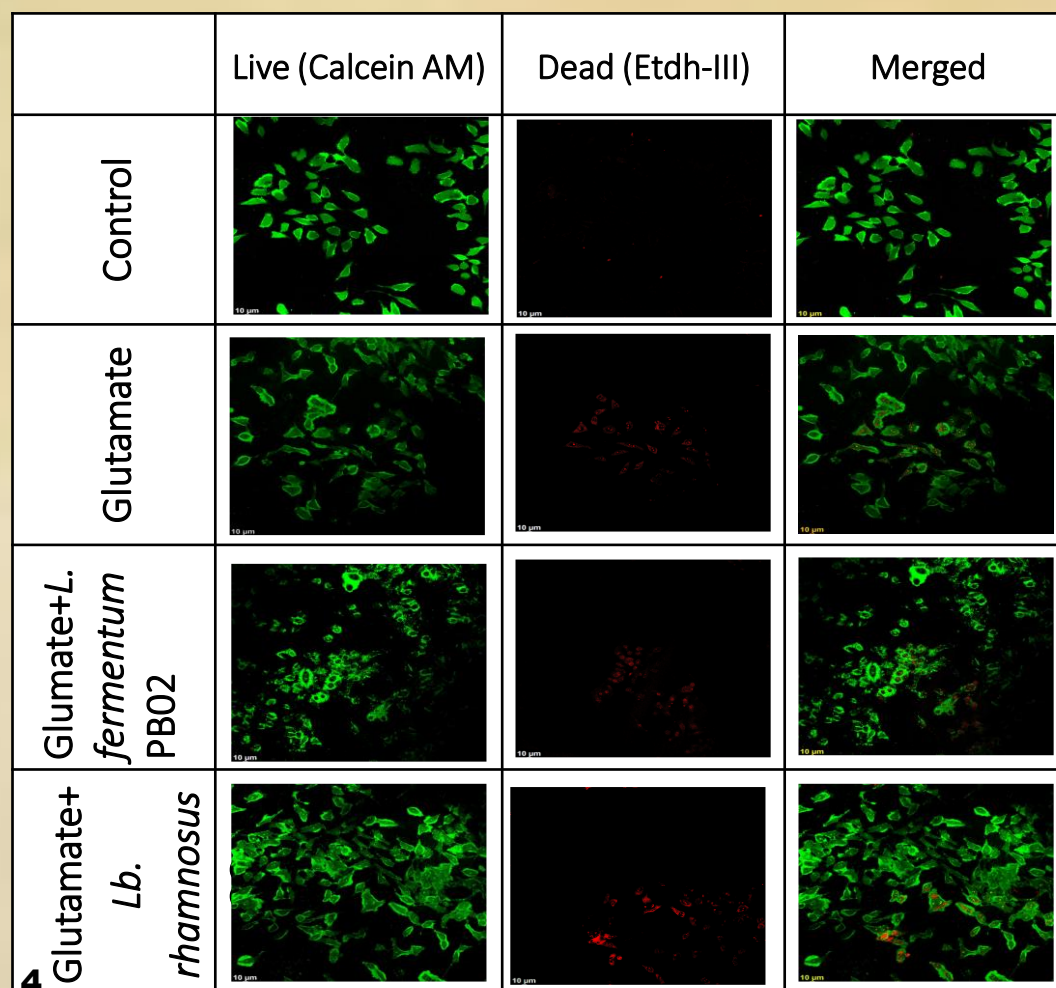
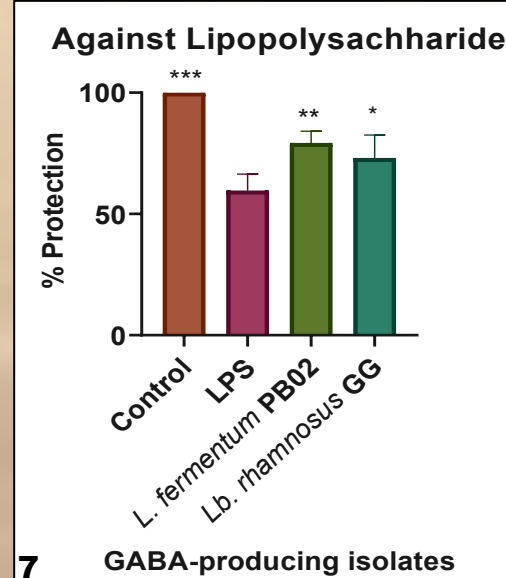
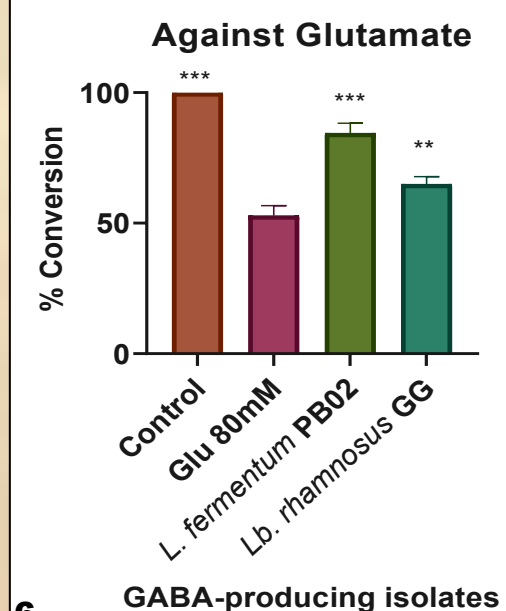
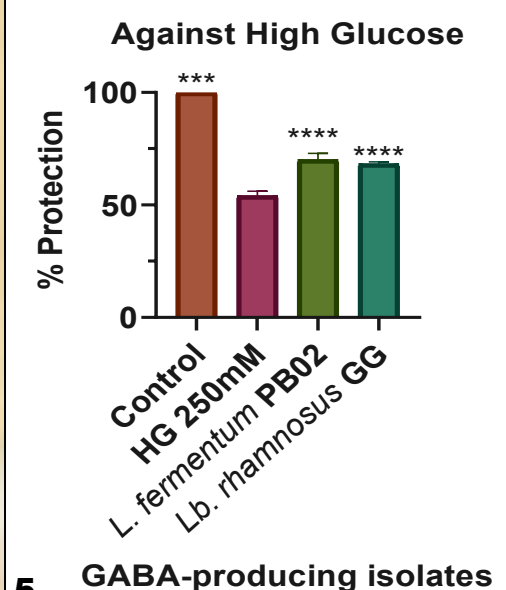


Figure 4: Confocal microscopic images -Live/Dead analysis

Conclusion

- Limosilactobacillus fermentum* PB02 (GenBank ID PQ555695) can potentially reduce glutamate toxicity by increasing GABA levels, modulating GABAergic signalling, and influencing the Gut-Brain Axis.
- The isolate showed a promising $79.2 \pm 4.87\%$ protection against LPS, $70.1 \pm 2.77\%$ protection against glucose, and could reverse glutamate toxicity by $84.47 \pm 3.86\%$ in the SH-SY5Y neural cell line model
- The strain offers moderate to high neuroprotection and could be explored as a potential psychobiotic after evaluating its neuroprotective effects in a depression-induced mouse model.
- Isolated strains belong to the phylum Bacillota (previously Firmicutes), and their desirable probiotic attributes make them a comparable candidate to existing probiotics.



Figures 5, 6, 7: Plots showing neuroprotection against different stress conditions