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ENHANCING THE BIOCONTROL POTENTIAL OF BACILLUS AMYLOLIQUEFACIENS BY HYDROGEL ENCAPSULATION FOR COMBATING INTESTINAL PATHOGENS

K Abirami * R Poojasri

Department of Microbiology

SRM Arts and ScienceCcollege Kattankulathur - 603203 Tamil Nadu- India

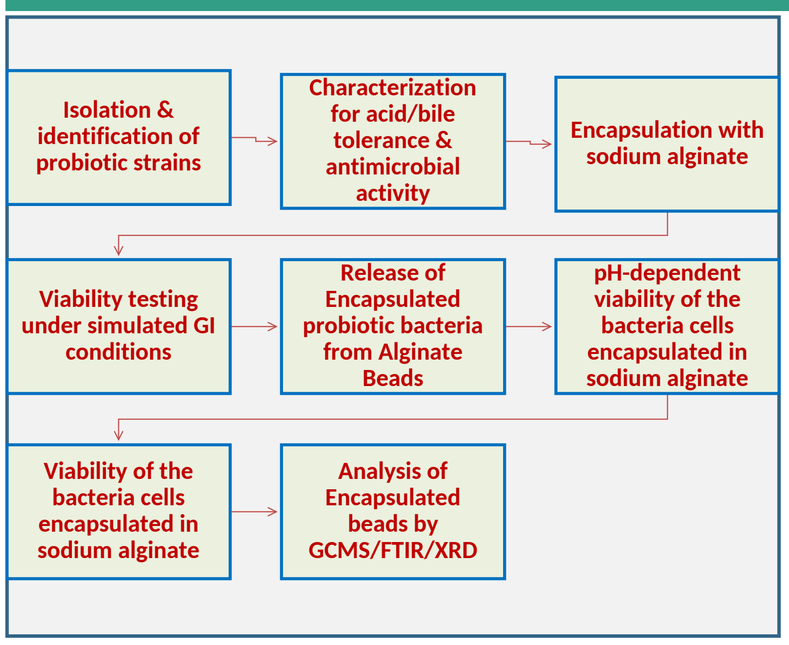
INTRODUCTION & AIM

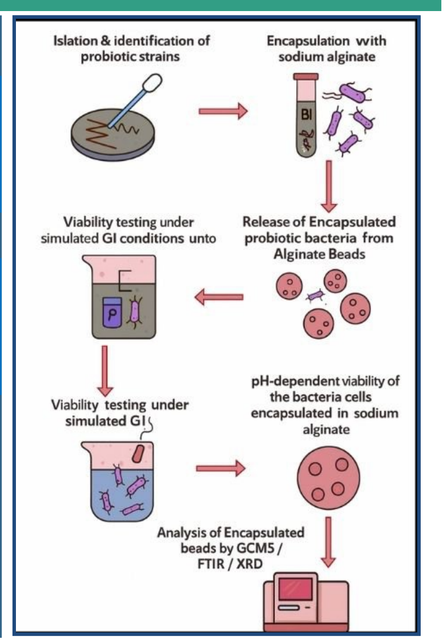
Microencapsulation surrounds solid, liquid, or gaseous cores with a polymeric shell to produce microcapsules ranging from a few micrometers to several millimeters in diameter. These capsules consist of a core (fill) and a wall (shell), which protects sensitive materials from environmental interactions. Applications span across the food, pharmaceutical, agricultural, and graphics industries. Functions of microencapsulation in foods (Shahidi & Han, 1993):

- ❖ Reduce reactivity of core materials
- ❖ Slow diffusion of active components
- ❖ Improve handling
- ❖ Control release behavior
- ❖ Mask undesirable flavors
- ❖ Allow dilution of potent ingredients

In pharmaceuticals, microencapsulation improves stability, masks taste, removes incompatibilities, and enables controlled or sustained drug release.

METHOD





RESULTS & DISCUSSION

- *Bacillus amyloliquefaciens* isolated from pearl millet was evaluated for probiotic potential through standard assays, including acid tolerance, hemolytic activity, cell surface hydrophobicity, auto-aggregation, NaCl tolerance, and phenol tolerance. The isolate exhibited positive results in all tests, confirming its probiotic attributes.
- To enhance stability and delivery, the strain was encapsulated in a sodium alginate hydrogel matrix. FTIR analysis verified successful encapsulation by identifying characteristic functional groups associated with alginate–cell interactions.

S.NO	MORPHOLOGICAL CHARACTERIZATION	OBSERVATION
1	Size	2mm
	Shape	Irregular
3	Pigmentation/ Colour	White
4	Margin	Lobate
5	Appearance	Wrinkle, Dry, Dull
6	Gram Staining	Gram positive rod
7	Motility	Non Motile

S.NO	BIOCHEMICAL TEST	RESULT
1	Indole	-
2	Methyl red	-
3	Voges Proskauer	+
4	Citrate Utilization	+
5	Triple Sugar Iron	+
6	Catalase	+

Table.1 Morphological characterization of the isolate and Fig.1 Physiological and biochemical properties of the isolate..

Fig.2 Phylogenetic analysis based on 16S rRNA gene sequencing

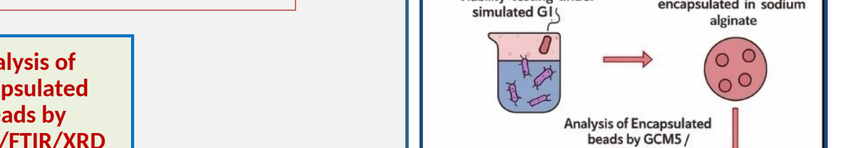


Fig3.Characterization of *B. amyloliquefaciens (a)* Acid tolerance test (b) Nacl tolerance test (c) Phenol tolerance test

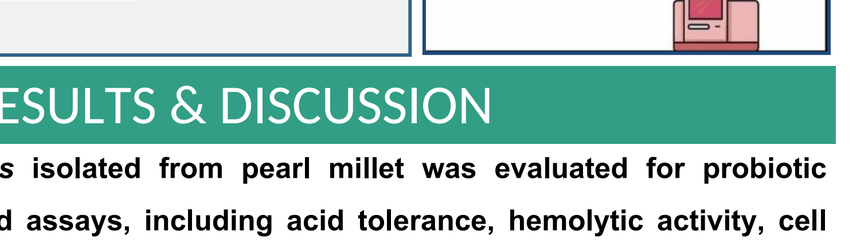


Figure 4 – (a) *B. amyloliquefaciens* trapped in sodium alginate beads.
(b) *B. amyloliquefaciens* trapped in sodium alginate beads.
(c) Viability assessment of encapsulated *B. amyloliquefaciens*

- XRD analysis showed a short-range diffraction peak at 2θ = 11.848°, indicating an amorphous encapsulated structure with a small crystalline lattice, supporting efficient entrapment of cells within the matrix.
- GC-MS metabolic profiling of the encapsulated strain identified 65 bioactive metabolites. Among them, 1,2-Benzenedicarboxylic acid, diethyl ester emerged as a key metabolite, suggesting its importance in bacterial metabolic activity under encapsulated conditions.
- Viability studies under acidic pH demonstrated high survival rates, confirming that encapsulation effectively protects the probiotic during gastrointestinal conditions.
- The cell-free supernatant of encapsulated *B. amyloliquefaciens* showed strong antibacterial activity, with inhibition zones of 14 mm (125 µL) against *Escherichia coli* and 17 mm (100 µL) against *Shigella*, validating its potential application in food and pharmaceutical sectors as a functional probiotic. agent.

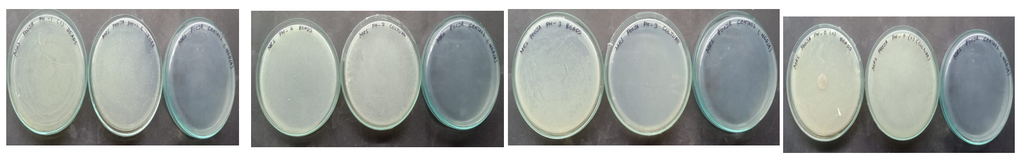


Figure 5 – (a) plate containing pH (pH – 1) adjusted free and encapsulated cells of *B. amyloliquefaciens.*
(b)plate containing pH (pH – 2) adjusted free and encapsulated cells of *B. amyloliquefaciens*
(c) plate containing pH (pH – 3) adjusted free and encapsulated cells of *B. amyloliquefaciens*
(d) plate containing unadjusted free and encapsulated cells of *B. amyloliquefaciens*

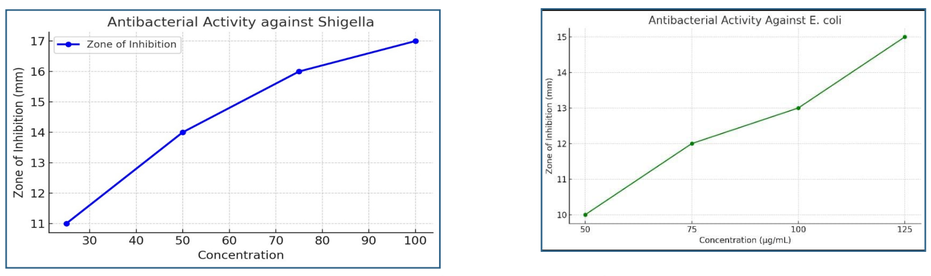


Figure 6 (a) Antibacterial activity *B. amyloliquefaciens* against *Shigella*
(b) Antibacterial activity *B. amyloliquefaciens* against *E.coli*

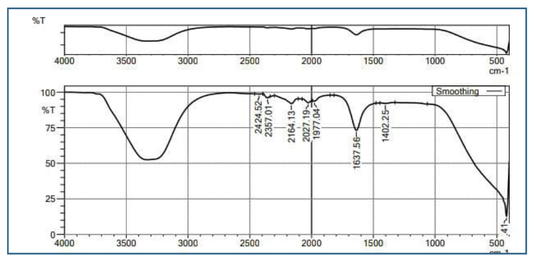


Figure 7 – FTIR analysis

S. No	PEAK VALVES	FUNCTIONAL GROUPS
01	422.41	C-I
02	1406.11	CH_3
03	1637.56	C=O
04	1977.04	C=O
05	2424.52	O-H
06	2424.52	O-H

Table 2 – Functional groups of Encapsulated bead supernatant

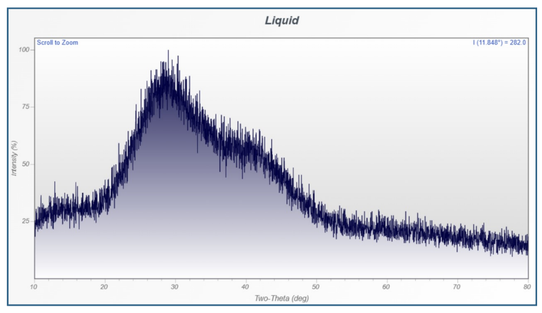


Figure 8 – XRD image of supernatant of encapsulated beads

CONCLUSION

Sodium alginate microencapsulation significantly improves probiotic stability, viability, and targeted release in the gastrointestinal tract. Coupling encapsulated probiotics with nutrient-rich pearl millet provides promising opportunities for functional foods, nutraceuticals, and therapeutic applications.

FUTURE WORK / REFERENCES

- ❖ Conduct advanced in vitro GI tract simulations (TIM-1, SHIME models) to evaluate release kinetics.
- ❖ Perform animal or human trials to determine colonization, metabolic effects, and overall health benefits.

❖ Afzaal et al. (2020); Fontana et al. (2013); Jyothi et al. (2012); Kaur et al. (2021); Object et al. (2018); Poshadri & Kuna (2010); Shahidi & Han (1993); Soccol et al. (2010); Wais et al. (2016)