Title: Preparation and characterization of chitosan nanocapsules containing the postbiotic Lactiplantibacillus plantarum and investigation of its antimicrobial and antioxidant effects

Abstract:

This study aimed to synthesize and characterize chitosan nanoparticles loaded with Lactiplantibacillus plantarum postbiotic and to evaluate their antioxidant, antibacterial, and antifungal activities. The nanoparticles were prepared via ionic gelation, and their physicochemical properties were analyzed using DLS, zeta potential, and SEM. The average particle size of blank chitosan was 140.1 nm, while postbiotic-loaded chitosan nanoparticles measured 206.0 nm, with zeta potentials of +17.1 and +60.4 mV, respectively, indicating good colloidal stability. SEM images confirmed spherical and uniform morphology, and FTIR spectra indicated molecular interactions between chitosan and the postbiotic.

Antioxidant activity assessed via DPPH and ABTS assays demonstrated that postbiotic-loaded nanoparticles exhibited the highest radical scavenging potential. At 20 mg/mL, DPPH inhibition was 48.92% and ABTS inhibition was 96.48%, significantly higher than postbiotic alone (DPPH: 51.73%, ABTS: 75.45%) and blank chitosan (DPPH: 62.39%, ABTS: 77.44%).

In antibacterial assays, postbiotic-loaded nanoparticles showed the strongest inhibition against *Escherichia coli*, *Staphylococcus aureus*, *Salmonella typhimurium*, and *Listeria monocytogenes*. At 100 mg/mL, the inhibition zone for *L. monocytogenes* was $24.25 \pm 0.17 \text{ mm}$. MIC and MBC values were significantly lower than those of postbiotic or blank chitosan, reflecting an approximately eightfold enhancement in antimicrobial efficacy.

For antifungal activity, postbiotic-loaded nanoparticles displayed superior inhibition against *Aspergillus niger* and *A. flavus*, whereas blank chitosan showed no inhibitory effect. At 40% concentration, the inhibition zones for *A. niger* and *A. flavus* were 18.40 mm and 17.31 mm, respectively, exceeding the effects of postbiotic alone.

Overall, encapsulating postbiotics into chitosan nanoparticles enhances physicochemical stability and significantly improves antioxidant, antibacterial, and antifungal activities, indicating their potential as promising bioactive agents in food and pharmaceutical applications.

Keywords: Postbiotic, Chitosan, Lactobacillus plantarum, Cell-free supernatant, Nanoparticle