

Microencapsulation of Probiotic and Evaluation for Viability, Antimicrobial Property and Cytotoxic Activities of its Postbiotic Metabolites on MCF-7 Breast Cancer Cell Line

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Abstract : Background: Probiotics are live microbial feed supplement beneficial for host. Probiotics and their postbiotic products have been used to prevent or treat various health conditions. However, the products cell viability is often low due to harsh conditions subjected during processing, handling, storage, and gastrointestinal transit. These strongly influence probiotics' benefits; thus, viability is essential for probiotics to produce health benefits for the host. Microencapsulation is a promising technique with considerable effects on probiotic survival. The study is aimed to formulate a microencapsulated probiotic and evaluate its viability, antimicrobial efficacy, and cytotoxic activity of its postbiotic on the MCF-7 breast cancer cell line. Method: Human and animal raw milk were sampled for lactic acid bacteria. The isolated bacteria were identified using conventional and VITEK 2 systems. The identified lactic acid bacterium was encapsulated using spray-dried and extrusion methods. The free, encapsulated, and chitosan-coated encapsulated probiotics were tested for viability in simulated-gastric intestinal (SGI) fluid and different storage conditions at refrigerated (4oC) and room (25oC) temperatures. The disintegration time and weight uniformity of the spray-dried hard gelatin capsules were tested. The antimicrobial property of free and encapsulated probiotics was tested against enteric pathogenic isolates from antiretroviral therapy (ART) treated HIV-positive patients. The postbiotic of the free cells was extracted, and its cytotoxic effect on the MCF-7 breast cancer cell line was tested through an MTT assay. Result: The *Lactobacillus plantarum* was isolated from animal raw milk. Zero-size hard gelatin L. *plantarum* capsules with granules within a size range of 0.71-1.00 mm diameter was formulated. The disintegration time ranges from 2.14 ± 0.045 to 2.91 ± 0.293 minutes, while the average weight is 502.1mg. Simulated gastric solution significantly affected viability of both free and microcapsules. However, the encapsulated cells were more protected and viable due to impermeability in the microcapsules. Furthermore, the viability of free cells stored at 4oC and 25oC were less than 4 log CFU/g and 6 log CFU/g respectively after 12 weeks. However, the microcapsules stored at 4oC achieved the highest viability among the free and microcapsules stored at 25oC and the free cells stored at 4oC. Encapsulated cells were released in the simulated gastric fluid, viable and effective against the enteric pathogens tested. However, chitosan-coated calcium alginate encapsulated probiotics significantly inhibited *Shigella flexneri*, *Candida albicans*, and *Escherichia coli*. The Postbiotic Metabolites (PM) of *L. plantarum* produced a cytotoxic effect on the MCF-7 breast cancer cell line. The postbiotic showed significant cytotoxic activity similar to 5FU, a standard antineoplastic agent. The inhibition concentration of 50% growth (IC50) of postbiotic metabolite K3 is low and consistent with the IC50 of the positive control (Cisplatin). Conclusions: *Lactobacillus plantarum* postbiotic exhibited a cytotoxic effect on the MCF-7 breast cancer cell line and could be used as combined adjuvant therapy in breast cancer management. The microencapsulation technique protects the probiotics, improving their viability and delivery to the gastrointestinal tract. Chitosan enhances antibacterial efficacy; thus, chitosan-coated microencapsulated *L. plantarum* probiotics could be more effective and used as a combined therapy in HIV management of opportunistic enteric infection.

Keywords : probiotics, encapsulation, gastrointestinal conditions, antimicrobial effect, postbiotic, cytotoxicity effect

Conference Title : ICMPPA 2023 : International Conference on Marine Probiotics, Prebiotics and their Applications

Conference Location : Rome, Italy

Conference Dates : November 20-21, 2023