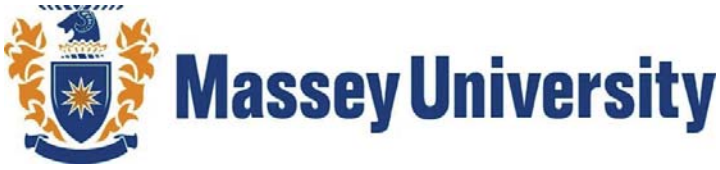
MAXIMIZING VIABILITY OF *LACTOBACILLUS PARACASEI* SUBSP. *PARACASEI L. CASEI* 431 DURING PROCESSING AND AMBIENT STORAGE



A THESIS PRESENTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF MASTER OF TECHNOLOGY IN FOOD TECHNOLOGY

BY

NAMRATA TANEJA

RIDDET INSTITUTE

MASSEY UNIVERSITY, PALMERSTON NORTH

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ABSTRACT

In the present study, fluidized bed drying has been examined as a low-energy alternative to more expensive freeze-drying of the probiotic commercial strain *Lactobacillus paracasei* subsp. *paracasei L. casei* 431. The aim of this study was to maximize the viability of *L. casei* 431 during laboratory and industrial scale processing and storage. The study proceeded in three stages: a) Optimizing the growth conditions and medium composition for maximizing cell growth and desiccation tolerance b) Standardizing the harvesting conditions (harvesting time and techniques) and mixing conditions (mixing of cells with protective carrier) to minimize the mixing and drying loss c) Investigating the effect of various parameters during fluidized bed (FB) drying (initial moisture content, drying time and drying temperature) through Plackett-Burman (PB) and factorial design with the objective of identifying the ideal combination for maximizing the viability of *L. casei* 431 during drying and ambient storage.

The preliminary experiments were performed to optimize growth medium composition under controlled pH in a bioreactor. The effect of supplementing de Man, Rogosa and Sharpe (MRS) media with glucose and yeast extract on viable cell count during batch and fed-batch fermentation was compared. The pH controlled fed-batch fermentation resulted in a 5 fold increase in the viable cell count when compared to batch fermentation. But when the cells obtained from this pH controlled media showed huge drying and storage losses as compared to the uncontrolled pH media, a sequential PB design followed by central composite design matrix was used to screen and optimize the factors that could maximize cell growth under uncontrolled pH conditions. The supplementation of yeast extract and meat extract at a high concentration of 0.6-0.8% nitrogen in MRS media increased the viable cells of *L. casei* 431 by more than 2 fold and biomass by more than 1.5 fold as compared to control (MRS media). The cells from uncontrolled pH fermentations were then harvested by high speed centrifugation and collected cells were mixed with protective carrier (whole milk powder) of different water activity under different mixing conditions. Once growth and mixing conditions were standardized, another PB design followed by factorial design was used to illustrate the effect of various parameters such as harvesting at different growth phases, total solids of harvested cells, initial moisture content, and drying conditions such as drying temperature and drying time, on residual moisture content in combination with their effect on drying and storage stability of *L. casei* 431 under ambient and accelerated storage (37 °C) conditions.

The results showed that the pH during growth, the growth phase, the harvested cell solids/moisture content, the mixing techniques, the drying temperature and the final moisture content were important factors affecting cell stability during drying and storage. Fermentation with acid stress and controlled fluidized bed drying were able to keep the *L. casei* 431 cells relatively stable for 3 months at 25 °C in heat sealed aluminum bags (with inner polyethylene layer) during laboratory and pilot/industrial scale preservation. It was further observed that the shelf-life of FB dried *L. casei* 431 cells at 25 ˚C was 6 times longer than at 37 ˚C.

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