

**IMPACT OF IN VITRO FERMENTATION, ENZYMATIC HYDROLYSIS, AND DIGESTION ON THE DEGRADATION OF B-CASOMORPHIN-7 IN MILK USING *LACTICASEIBACILLUS CASEI* AND *LIMOSILACTOBACILLUS FERMENTUM***

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The objective of this study was to reduce the formation of  $\beta$ -casomorphin-7 (BCM-7), an opioid peptide derived from  $\beta$ -casein A1, associated with inflammation and gastrointestinal disorders. To achieve this, strategies involving thermal treatments and bacterial fermentation with *Lacticaseibacillus casei* and *Limosilactobacillus fermentum*, combined with enzymatic hydrolysis, were evaluated. The thermal treatment at  $\beta$ -casomorphin-7 100°C for 30 minutes increased the concentration of soluble proteins to  $7.58 \pm 0.07$  mg/mL, while autoclaving at 105°C for 15 minutes had a lesser impact, resulting in  $3.24 \pm 0.05$  mg/mL, this method was chosen as the standard to minimize protein degradation and prepare the milk for subsequent processes. Bacterial fermentation preserved the proteins during digestion, while the control showed a significant drop in soluble protein concentrations, from  $5.32 \pm 0.05$  mg/mL to  $0.19 \pm 0.01$  mg/mL. SDS-PAGE evidenced the degradation of milk proteins, including  $\beta$ -casein. The ELISA showed that *L. fermentum* was more efficient in reducing BCM-7, decreasing its concentration from  $0.22 \pm 0.01$  to  $0.08 \pm 0.01$ . The combination of *L. casei* and *L. fermentum* did not result in any additional significant reduction. Thus, fermentation combined with enzymatic hydrolysis proved effective in reducing the formation of BCM-7.

**Keywords:** BCM-7. Fermentation. Enzymatic Hydrolysis.