Thesis Title	Effects of Lactic Acid Fermentation on Citrulline Content in Watermelon (*Citrullus lanatus*) Rind
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ABSTRACT

Watermelon rind, a major agricultural by-product of watermelon, is often discarded as waste despite its edibility. Fermentation *in situ* offers a sustainable approach to reintegrating such underutilized materials into the food value chain by enhancing preservation, safety, and bioactive compounds. This study aimed to optimize fermentation conditions to enhance L-citrulline, the predominant functional amino acid in watermelon rind with known health benefits. Natural fermentation was carried out by varying salt concentration (2%, 4%, and 6%), temperature (25, 30, 35, and 40 °C), and duration, and their effects on microbial dynamics, acidification, amino acid profile including citrulline and its metabolic flux, and other phytochemical properties were evaluated. Microbiological analysis demonstrated that 6% salt facilitated the highest lactic acid bacteria (LAB) growth, reaching peak populations across all temperatures. At 30°C, sustained LAB growth was observed with the minimum final pH (3.23±0.01), establishing it as the optimal temperature for controlled fermentation. Enterobacteriaceae, initially present at high levels (7.35–7.72 log CFU/g), became undetectable in all samples by day 5, while yeast and mold populations were suppressed within 2 to 6 days. Subsequent fermentation at 30°C for 48 h demonstrated that amino acids involved in the arginine deiminase (ADI) pathway were strongly influenced by both salt concentration and fermentation time. While low-salt conditions (2% and 4%) led to citrulline depletion through its conversion to ornithine, 6% salt inhibited subsequent citrulline metabolism, resulting in significant accumulation reaching 737.12±12.50 mg/kg at 12 hours, compared to 652.41±7.77 mg/kg in the unfermented sample. Although citrulline levels declined thereafter, they remained significantly higher than the initial concentration throughout fermentation. These findings indicate that fermentation for 12 h at 6% salt concentration represents

the optimal condition for enhancing citrulline content. Total phenolic content (TPC) increased significantly across all treatments, with the 2% salt sample showing the highest relative increment (71.59%). DPPH radical scavenging activity more than doubled after 48 h, reaching 1.189–1.281 mg TE/g FW. Among free amino acids, glutamic acid and aspartic acid were more abundant at lower salt levels, suggesting greater umami character, while proline, threonine, histidine, lysine, and glutamine were elevated at higher salt concentrations. In case of color parameters, 6% salt better preserved rind lightness and yellowness, although greenness declined across treatments due to acid-induced chlorophyll degradation. These findings demonstrate that higher salt concentration not only promotes higher LAB counts but also drives citrulline overproduction, particularly at short fermentation durations. These findings support the valorization of watermelon rind by defining optimized fermentation conditions for citrulline enhancement, contributing to both agricultural waste reduction and the development of functional fermented products.

Keywords: Watermelon Rind, Fermentation, Citrulline, Lactic Acid Bacteria, Valorization, Amino Deiminase Pathway