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Content

INFLUENCE OF AMINO ACID SUBSTITUTION ON THE ANTIMICROBIAL ACTIVITY OF BACTERIOCIN LACTOLISTERIN BU

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Introduction: Lactolisterin BU (LBU) is a potent bacteriocin derived from *Lactococcus lactis* subsp. *lactis* bv. diacetylactis BGBU1-4. It exhibits antimicrobial properties against Gram-positive food spoilage and foodborne pathogens. This research aimed to explore the impact of amino acid substitution in LBU on its antimicrobial activity by utilizing *in silico* prediction of LBU's secondary structure and amino acid substitutions.

Methods: The secondary structure of LBU was predicted using Phyre2 software. Five variants of LBU were selected and chemically synthesized, along with unaltered LBU and BHT-B, serving as controls. Peptides were twofold diluted in distilled water, resulting in final concentrations ranging from 1000 µg/ml to 0.5 µg/ml. An agar spot test, employing 5 µl of the dilution, was conducted on three indicator strains: *Lactococcus lactis* BGMN1-596, *Listeria monocytogenes* ATCC19111, and *Staphylococcus aureus* ATCC25923. The presence of inhibition zones was analyzed after overnight incubation at 37°C (*S. aureus*) and 30°C (*L. lactis* and *L. monocytogenes*).

Results: Phyre2 analysis unveiled the presence of two α-helices in LBU's structure. The majority of LBU variants displayed altered antimicrobial activity, with some changes being genus specific, potentially attributable to variances in cell wall composition. Some variants completely lost their activity, underscoring the significance of native amino acids or their physicochemical properties in the corresponding positions within LBU's structure. Furthermore, it was confirmed that chemically synthesized LBU effectively retains its antimicrobial activity.

Conclusion: Changes in amino acid composition give insight on structure-function relationship of LBU.

Key words: peptides; antimicrobials; LAB, lactolisterin BU; amino acids

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