

MEMBRANE DISRUPTION ANTIMICROBIAL MECHANISM OF *CHANNA STRIATUS* LYSOZYME-DERIVED ANTIMICROBIAL PEPTIDES (AMP)

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ABSTRACT

The increase in multidrug resistant bacteria has sparked an interest in the development of novel antibiotics. Antimicrobial peptides (AMPs) that operate by crossing the cell membrane may also have the potential to deliver drugs to intracellular targets. Lysozyme is one of the key antimicrobial peptides that act on the bacterial membrane to destroy them. In this study, a goose type lysozyme (LyzG) cDNA sequence was identified from the transcriptome library of freshwater fish *Channa striatus* (Cs). The cDNA contains 585 nucleotides which encodes a protein of 194 amino acids. CsLyzG shared a high sequence identity with other goose type lysozyme. The described protein sequence contained a GEWL domain with conserved GLMQ motif, 7 active residues and 2 catalytic residues. Gene expression analysis was performed to investigate which organs of *C. striatus* expressed CsLysG. It was shown that CsLyzG was distributed in major immune organs with highest expression found in the head kidney. The time-dependent expression of CsLysG mRNA, following bacterial (*Aeromonas hydrophila*) and fungal (*Aphanomyces invadans*) challenges, indicated a differential temporal expression pattern of this gene, ensuring its constant antimicrobial activity against both pathogens. Based on the propensity scale, we have identified and synthesized a novel AMP, IK12 and compared its activity with a previously reported peptide, TS10. The results of the antibiogram with IK12 showed that it was active against *Salmonella enterica*, a major multi-drug resistant (MDR) bacterial pathogen. IK12 induced loss of cell viability in the bacterial pathogen. We established that IK12 uses a membrane disruption antimicrobial mechanism using flow cytometry and scanning electron microscope (SEM), with blebbing observed around the bacterial cell membrane. In conclusion, CsLyzG is a potential innate immune component, which can be efficiently used as an eco-friendly antibacterial substance in aquaculture which could be possibly delivered by peptide encapsulation technique using fish oil.

KEYWORDS

Lysozyme; Antimicrobial peptide; Membrane disruption; Gene expression; *Channa striatus*

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