World Academy of Science, Engineering and Technology
International Journal of Biomedical and Biological Engineering
Vol:19, No:09, 2025

Integrated Approach to Attenuate Insulin Amyloidosis: Synergistic Effects of Peptide and Cysteine Protease Enzymes

Authors : Shilpa Mukundaraj, Nagaraju Shivaiah

Abstract : Amyloidogenic conditions, driven by protein aggregation into insoluble fibrils, pose significant challenges in diabetes management, particularly through the amyloidogenic LVEALYL sequence in insulin B-chain. This study explores a dual therapeutic strategy involving cysteine protease enzymes and inhibitory peptides to target insulin amyloidosis. Combining in silico, in vitro, and in vivo methodologies, the research aims to inhibit amyloid formation and degrade preformed fibrils. Inhibitory peptides were designed using structure-guided approaches in Rosetta to specifically target the LVEALYL sequence. Concurrently, cysteine protease enzymes, including papain and ficin, were evaluated for their fibril disassembly potential. In vitro experiments utilizing SDS- PAGE and spectroscopic techniques confirmed dose-dependent degradation of amyloid aggregates by these enzymes, with significant disaggregation observed at higher concentrations. Peptide inhibitors effectively reduced fibril formation, as evidenced by reduced Thioflavin T fluorescence and circular dichroism spectroscopy. Complementary in silico analyses, including molecular docking and dynamic simulations, provided structural insights into enzyme binding interactions with amyloidogenic regions. Key residues involved in substrate recognition and cleavage were identified, with computational findings aligning strongly with experimental data. These insights confirmed the specificity of papain and ficin in targeting insulin fibrils. For translational potential, an in vivo rat model was developed involving subcutaneous insulin amyloid injections to induce localized amyloid deposits. Over six days of enzyme treatment, a marked reduction in amyloid burden was observed through histological and biochemical assays. Furthermore, inflammatory markers were significantly attenuated in treated groups, emphasizing the dual role of enzymes in amyloid clearance and inflammation modulation. This integrative study highlights the promise of cysteine protease enzymes and inhibitory peptides as complementary therapeutic strategies for managing insulin amyloidosis. By targeting both the formation and persistence of amyloid fibrils, this dual approach offers a novel and effective avenue for amyloidosis treatment.

Keywords : insulin amyloidosis, peptide inhibitors, cysteine protease enzymes, amyloid degradation
Conference Title : ICA 2025 : International Conference on Amyloidosis
Conference Location : Singapore, Singapore
Conference Dates : September 09-10, 2025

Open Science Index, Biomedical and Biological Engineering Vol:19, No:09, 2025 publications.waset.org/abstracts/196206.pdf