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Investigating the Factors Influencing Human Serum Albumin Fibrillation Using Thioflavin T

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Abstract

Protein fibrillation, a process implicated in various neurodegenerative diseases, is a critical area of study for understanding amyloid-related pathologies and developing therapeutic strategies. This study utilized human serum albumin (HSA), a well-characterized model protein, to explore how temperature, pH, and protein concentration affect fibrillation. A Central Composite Design (CCD) experimental framework was implemented, assessing three levels for each factor: temperature (37°C, 47°C, and 57°C), pH (3, 5, and 7), and concentration (1, 1.5, and 2 mg/mL). Fibrillation was induced over a 48-hour period at a stirring speed of 300 rpm, with aggregation monitored using thioflavin T (ThT), a fluorescent dye that selectively binds to amyloid fibrils, allowing real-time observation of the fibrillation dynamics. Fluorescence intensity was recorded at an excitation wavelength of 440 nm and emission wavelengths of 450 nm and 600 nm. Results indicated that the combined effects of temperature, pH, and concentration significantly influenced HSA fibrillation, with aggregation observed. The quadratic model accounted for 92% of the variance in yield, highlighting significant contributions from concentration. Normalization was performed using a power transformation of -1.24. The model demonstrated significance with a p-value of 0.0013 and an F-value of 10.85. The optimal conditions identified were a concentration of 2 mg/mL at 57°C and pH 3. This research enhances our understanding of the conditions that facilitate fibrillation.

Key words: Protein Fibrillation, Human Serum Albumin, Thioflavin T, amyloid-related diseases

References

[1] Schneider TR, Stöckli L, Felbecker A, Nirmalraj PN. Protein fibril aggregation on red blood cells: a potential biomarker to distinguish neurodegenerative diseases from healthy aging. *Brain Communications*; 6(3): fcae180, 2024.
[2] Wodrich M, et al. Editorial: Protein misfolding, altered mechanisms and neurodegeneration. *Frontiers in Molecular Neuroscience*; 2023; 16: 1134855.
[3] Wodrich M, et al. Editorial: Protein misfolding, altered mechanisms and neurodegeneration. *Frontiers in Molecular Neuroscience*; 2023; 16: 1134855.