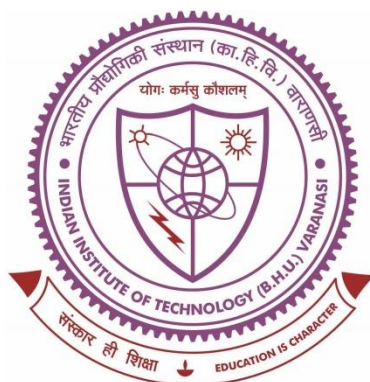


**Structure Activity Relationship Studies on Ferulic Acid  
Template-Based Novel Molecules to Improve Upon  
Multifunctional Properties for The Management of  
Alzheimer's Disease**



**Thesis submitted in partial fulfilment for the  
Award of Degree**

**Doctor of Philosophy**

**By**

***GOURAV SINGH***

**DEPARTMENT OF PHARMACEUTICAL ENGINEERING & TECHNOLOGY**

**INDIAN INSTITUTE OF TECHNOLOGY**

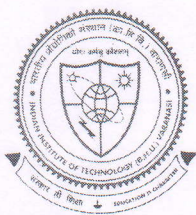
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I, **Gourav Singh**, certify that the work embodied in this Ph.D. thesis is my own bonafide work and carried out by me under the supervision of **Dr. Gyan Prakash Modi** from **January, 2019 to July, 2024** at the **Department of Pharmaceutical Engineering & Technology**, Indian Institute of Technology (B.H.U.), Varanasi. The matter embodied in this thesis has not been submitted for the award of any other degree/diploma.

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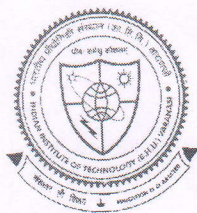
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## List of Abbreviations

| Abbreviations               | Full forms                                                                                                      |
|-----------------------------|-----------------------------------------------------------------------------------------------------------------|
| AD                          | Alzheimer's disease                                                                                             |
| ACh                         | Acetylcholine                                                                                                   |
| <i>h</i> ACh                | Human acetylcholine                                                                                             |
| MW                          | Molecular weight                                                                                                |
| MTT                         | 3- (4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium                                                           |
| PAS                         | Peripheral anionic site                                                                                         |
| ROS                         | Reactive oxygen species                                                                                         |
| TMS                         | Tetramethylsilane                                                                                               |
| BChE                        | Butyrylcholinesterase                                                                                           |
| BBB                         | Blood-Brain Barrier                                                                                             |
| CAS                         | Catalytic active site                                                                                           |
| CDCl <sub>3</sub>           | Deuterated chloroform                                                                                           |
| DMSO- <i>d</i> <sub>6</sub> | Deuterated dimethyl sulfoxide- <i>d</i> <sub>6</sub>                                                            |
| DPZ                         | Donepezil                                                                                                       |
| DTNB                        | 5,5'-dithiobis-2-nitrobenzoic acid                                                                              |
| DPPH                        | 2,2-diphenyl-1-picrylhydrazyl                                                                                   |
| EDCI.HCl                    | 1-[3-(dimethylamino)-propyl]-3-ethyl carbodiimide<br>hydrochloride                                              |
| FA                          | Ferulic acid                                                                                                    |
| MABs                        | Monoclonal antibodies                                                                                           |
| JNK                         | Janus kinase                                                                                                    |
| HRMS                        | High-resolution mass spectrometry                                                                               |
| HOBt                        | <i>N</i> -hydroxybenzotriazole                                                                                  |
| IC <sub>50</sub>            | The inhibitory concentration required to kill 50% of the<br>population                                          |
| IL-6                        | Interleukin-6                                                                                                   |
| IL-β                        | Interleukin-6                                                                                                   |
| MMP                         | Interleukin-beta                                                                                                |
| TLR4                        | Mitochondrial membrane potential                                                                                |
| NLRP3                       | Toll-like receptor 4<br>Nucleotide-binding domain, leucine-rich-containing family,<br>pyrin domain-containing-3 |
| CAT                         | Catalase                                                                                                        |
| MDA                         | Malondialdehyde                                                                                                 |
| SOD                         | Superoxide dismutase                                                                                            |

## List of Symbols

| Symbols            | Meaning                |
|--------------------|------------------------|
| $\alpha$           | Alpha                  |
| $\beta$            | Beta                   |
| $\delta$           | Delta                  |
| $^{\circ}\text{C}$ | Degree Celsius         |
| $\text{\AA}$       | Angstrom               |
| mg                 | Milligram              |
| $\mu\text{g}$      | Micro gram             |
| $\mu\text{M}$      | Micromole              |
| mmol               | Millimole              |
| mL                 | Milliliter             |
| $\mu\text{L}$      | Microliter             |
| h                  | Hour                   |
| s                  | Singlet                |
| nm                 | Nanometer              |
| $\mu\text{m}$      | Micrometer             |
| mm                 | Millimeter             |
| cm                 | Centimeter             |
| ppm                | Parts per million      |
| rpm                | Revolutions per minute |
| Kcal               | Kilocalories           |
| Hz                 | Hertz                  |
| MHz                | Megahertz              |
| $J$                | Coupling constant      |
| d                  | Doublet                |
| t                  | Triplet                |
| q                  | Quartet                |
| m                  | Multiplet              |
| dd                 | Doublet of doublet     |
| $m/z$              | Mass-to-charge ratio   |
| %                  | Percent                |
| pH                 | Potential of hydrogen  |
| $\leq$             | Less than or equal     |
| $<$                | Less than              |
| $>$                | More than              |
| $\pm$              | Plus or minus          |

## Preface

Alzheimer's Disease (AD) is an age-related neurodegenerative disorder that accounts for over 80% of dementia cases in older adults worldwide. It is characterized by low level of acetylcholine (ACh), Increase of oxidative stress, accumulation of metals, deposition of amyloid-beta ( $A\beta$ ) plaques and neurofibrillary tangles, leading to the progressive loss of memory and cognitive functions. Despite decades of research on the etiology of the disease and substantial efforts by the pharmaceutical industry to develop effective therapies, no treatment has been found to cure AD or significantly inhibit its progression. Currently, four drugs—donepezil, galantamine, rivastigmine (acting on the cholinergic pathway), and memantine (acting on the NMDA receptor) and are approved by the USFDA. Aducanumab and Lecanemab are monoclonal antibodies (MABs) recently approved for the treatment of AD. These drugs target amyloid-beta ( $A\beta$ ) plaques, which are believed to play a central role in the pathophysiology of AD. Given the complex and multifactorial nature of AD, developing multifunctional ligands is a more promising approach.

**The present study is divided into six chapters:**

**Chapter 1:** This chapter overviews Alzheimer's disease, including its pathophysiology and the current treatments.

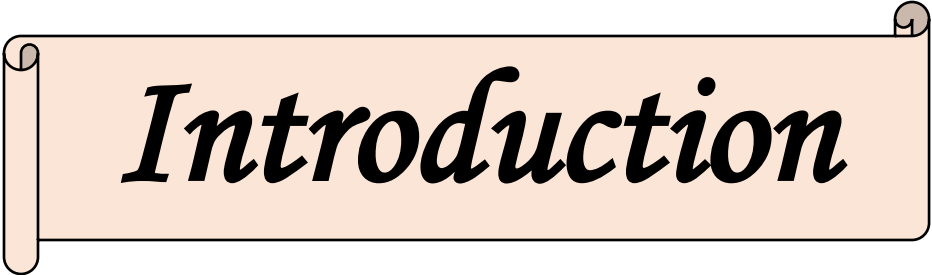
**Chapter 2:** This chapter presents a comprehensive review of the existing literature related to the relevant research.

**Chapter 3:** This chapter outlines the research work's hypothesis, rationale, and detailed plan.

**Chapter 4:** This chapter discusses the rationale for synthesizing and evaluating novel ferulic acid derivatives, specifically glycine/piperazine amide/benzylpiperazine/tryptamine derivatives. The designed molecules were synthesized and subjected to *in-vitro* enzyme inhibition studies. The potent molecules identified from these studies were further investigated for enzyme kinetics, antioxidant activity, metal chelation,  $A\beta$  modulation, and neuroprotection. Lead compounds were then selected for *in-vivo* studies in AD animal models to evaluate their effects on working memory and learning.

**Chapter 5:** This chapter details the general synthetic procedures used for the targeted compounds and their subsequent biological evaluation.

**Chapter 6:** This chapter provides the final summary and conclusions drawn from the research.



# *Introduction*