Design, Synthesis and Evaluation of Substituted Acetamides and Carbohydrazones as Dual Monoamine Oxidase and Acetylcholinesterase Inhibitors

Thesis submitted in partial fulfilment
for the award of degree

DOCTOR OF PHILOSOPHY

By

Mr. Sandeep Kumar

PHARMACEUTICAL ENGINEERING & TECHNOLOGY
INDIAN INSTITUTE OF TECHNOLOGY (BHU)
VARANASI – 221 005, UP
INDIA

Roll No.: 18161511

Year: 2024

PHARMACEUTICAL ENGINEERING & TECHNOLOGY

भारतीय प्रौद्योगिकी संस्थान IIT INDIAN INSTITUTE OF TECHNOLOGY BANARAS HINDU UNIVERSITY

Senthil Raja A, Professor

25 October 2024

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It is certified that the work contained in the thesis titled "**Design, Synthesis and Evaluation of Substituted Acetamidees and Carbohydrazones as Dual Monoamine Oxidase and Acetylcholinesterase Inhibitors**" by **Mr. Sandeep Kumar** has been carried out under my supervision and that this work has not been submitted elsewhere for a degree.

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Supervisor

25/10/24

(Senthil Raja A)

Professor (Pharmaceutical Chemistry),
Pharmaceutical Engineering and Technology
IIT (BHU) Varanasi-221005

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I, **Sandeep Kumar** certify that the work embodied in this PhD thesis is my own bona fide work and carried out by me under the supervision of **Prof. Senthil Raja A**, from July 2019 to October 2024, at the Department of Pharmaceutical Engineering and Technology, Indian Institute of Technology (Banaras Hindu University), Varanasi. The matter embodied in this thesis has not been submitted for the award of any other degree/diploma. I declare that I have faithfully acknowledged and given credits to the research workers wherever their works have been cited in my work in this thesis. I further declare that I have not wilfully copied any other's work, paragraphs, text, data, results, *etc.*, reported in journals, books, magazines, dissertations, theses, *etc.*, or available at websites and have not included them in this PhD thesis and have not cited as my own work.

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(Dr. Senthil Raja A)

Supervisor

S. Hemalatha 25/10/24.

(Dr. S. Hemalatha)

Head of the Department
विभागाध्यक्ष / Head
भैषजकीय अभियांत्रिकी एवं प्रौद्योगिकी विभाग /
Department of Pharmaceutical Engineering & Technology
भारतीय प्रौद्योगिकी संस्थान / INDIAN INSTITUTE OF TECHNOLOGY
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Sandeep Kumar

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF SCHEMES

LIST OF TABLES

LIST OF ABBREVIATIONS

2D	Two dimensional
3D	Three dimensional
Aβ	Amyloid beta
ACh	Acetylcholine
AChE	Acetylcholinesterase
AD	Alzheimer's disease
ADME	Absorption, distribution, metabolism, and excretion
ALS	Amyotrophic lateral sclerosis
ATCI	Acetylthiocholine iodide
BBB	Blood-brain barrier
BTCI	Butyrylthiocholine iodide
CAS	Catalytic anionic site
$CDCl_3$	Deuterated chloroform
DMF	N,N-dimethylformamide
DMSO-d_6	Deuterated dimethyl sulfoxide
DPZ	Donepezil
DPPH	2,2-Dithphenyl-1-picrylhydrazyl
DTNB	5,5'-Dithiobis-2-nitrobenzoic acid
*ee*AChE	Electric eel acetylcholinesterase
*h*AChE	Human acetylcholinesterase
HBA	Hydrogen bond acceptor
HBD	Hydrogen bond donor
HRMS	High resolution mass spectrometry
MTT	3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium
MTDL	Multitarget directed ligand
PAS	Peripheral anionic site
PDB	Protein data bank
PBL	Porcine brain lipid
SAR	Structure activity relationship
SEM	Standard error of mean
TMS	Tetramethylsilane

LIST OF SYMBOLS

α	Alpha
β	Beta
Δ	Delta
λ	Lambda
ºC	Degree Celsius
Å	Angstrom
mg	Milligram
µg	Microgram
µM	Micromolar
mM	Millimolar
mL	Millilitre
µL	Microlitre
h	Hour
nm	Nanometre
ppm	Parts per million
rpm	Revolutions per minute
kcal	Kilocalories
MHz	Megahertz
J	Coupling constant
d	Doublet
t	Triplet
m	Multiplet
dd	Doublet of doublet
m/z	Mass-to-charge ratio
%	Percent
pH	Potential of hydrogen
<	Less than
≤	Less than, or equal
>	More than
±	Plus, or minus
~	Approximate

PREFACE

The evolving complex pathophysiology of neurodegenerative diseases (NDDs) poses a big challenge to medicinal chemists to discover potential small molecule therapies. The available anti-neurodegenerative therapies provide only a symptomatic relief to patients and lack the desired efficacy; therefore, effective therapeutics are needed. Multitarget-directed ligands (MTDLs) capable of inhibiting brain monoamine oxidase-B (MAO-B) and acetylcholinesterase (AChE) and chelating brain iron ions have significant advantages in treating NDDs. Several potential MTDLs have been explored, however their further clinical development is limited by their poor neuroprotective properties *in vivo*. Thus, there is a need to develop efficacious MTDLs possessing a balanced multifunctional profile. In the current study, we have employed pharmacophore-based virtual screening and ligand-guided optimization approaches to develop new MTDLs possessing dual MAO-B-AChE inhibition, iron chelation and neuroprotective properties.

The present study is divided into five chapters and are as follows.

Chapter 1 describes the background, pathophysiology, and available drug therapies for NDDs and provides an exhaustive literature review on the reported dual MAO-ChE inhibitory hybrids/scaffolds followed by the research rationale and objectives of the study.

Chapter 2 deals with virtual screening-guided design, synthesis, characterization and biological evaluation of benzothiazole-derived thioacetamides as potential MTDLs (**ZBTI series**).

Chapter 3 deals with the design, synthesis, and biological evaluation of sesamol-derived O-acetamides (**SMA series**) and sesamol-derived extended carbohydrazones (**SMH series**).

Chapter 4 documents the design, synthesis, characterization and biological evaluation of eugenol/isoeugenol-derived carbohydrazones (**SEH & SIEH series**).

Chapter 5 presents the concluding remarks and future scope.

At the end, an appendix containing supporting information, spectral data of representative compounds, and a list of publications is included.