Abstract

Alzheimer's Disease (AD) is most common neurodegenerative disorder, which accounts for more than 80% of dementia cases worldwide in older people. It is characterized by deposition of amyloid β and neuro-fibrillary tangles. The disease leads to progressive loss of memory, functional ability to learn and is primarily characterized by the progressive loss of memory associated with other cognitive deficits. The global burden on the population suffering from AD was assessed to 44 million in 2015. This number is expected to double by 2030 and triple by 2050, if no effective treatment is in place. In a country like India, where approximately 41 % of the population is in the age group of 25 to 55 years, this needs to be seriously acknowledged and addressed.

Over the last few years, several targets of the disease have been identified. This include, but not limited to, Amyloid-β, Acetylcholinesterase (AChE), Butyrylcholinesterase (BuChE), β-secretase, γ-secretase, β-site APP cleaving enzyme-1 (BACE-1), NMDA receptors, Matrix metalloproteinases (MMPs), Neurofibrillary tangles *etc*.

Despite decades of study on the etiology of disease and also significant efforts by pharmaceutical industry to develop therapies, there is no effective treatment available to cure AD or to inhibit significantly its progression. However, there are four drugs viz. donepezil, galantamine, and rivastigmine, approved by USFDA, acting on cholinergic pathway and recently (2003) approved drug Memantine acting on NMDA receptor.

Considering the complex and multifactorial nature of disease, development of multifunctional ligands was considered a better option.

The present study is being divided into eight chapters. Chapter 1 deals with different types of neurodegenerative disorders, pathophysiology of AD and available treatments. Chapter 2 provides insight in the literature reports related to the work from scholarly

articles which forms the basis of the study. The section summarizes drugable targets for the treatment of AD and agonists or antagonists developed in the recent times. The mechanism of action of inhibitors and the molecular features of the receptors are also discussed in detail. The structural requirements for the agonistic or antagonistic action are explained along with structure-activity relationship (SAR) of the molecules. Chapter 3 includes the hypothesis of the study, and rationale of the work. It also incorporates the plan of study that helped to accomplish the research envisaged.

Chapter 4 deals with the rationale for syntheses of novel piperazinediones, obtained from *in-silico* studies and data mining. The development of pharmacophore was based on the hybrid drug design considering donepezil and PQ912, a chemical moiety in phase II of clinical trial. The designed hybrid pharmacophore was used for extensive data mining by using zinc and asinex databank which yielded over thousand compounds having desired pharmacophoric pattern. These data were further subjected to different filters viz. docking, *in-silico* BBB permeability and finally toxicity filter. The fully optimized molecules thus obtained were promoted to synthesis, *in-vitro* enzyme assays and *in-vivo* studies.. The potent molecules obtained from *in-vitro* study were further investigated for neuroprotection ability in MC 65 cells and antioxidant assay. Most potent compounds were selected for *in-vivo* studies in AD animal models to evaluate the working memory and learning response.

A few, piperazinediones developed also showed anxiolytic property which was further assessed and constitute chapter 5 *i.e.* biological profiling of piperazinediones for the management of anxiety associated with AD. In this, behavioral study on animal model was performed which was followed by estimation of neurotransmitter level in brain. The detailed mode of action of compounds was also assessed by Flumazenil antagonism on anxiolytic activity of compound.

In the next chapter (Chapter 6), synthesis of adamantly analogous as NMDA antagonists is includes. Memantine, an approved NMDA antagonist, was retained in the final structure and further optimization was done using the fragments of hits obtained from virtual screening. Triazole moiety, reported to exhibit neuroprotective effect was incorporated in some of the compounds. The synthesized compounds were subjected to *in-vitro* MMP-2 inhibition assay, inhibition assay of metal-induced $A\beta_{1-42}$ aggregation, confocal fluorescence imaging, antioxidant activity (DPPH assay), MC65 neuroprotection assay, and electrophysiology on different glutamate and glycine-mediated NMDA receptors and results were analyzed.

Another series of novel compounds synthesized, are presented in chapter 7. This includes quinoline analogues as potent inhibitors of AChE, BuChE, and BACE-1. Fragments from different bioactive molecules *viz* donepezil, LY2811376, MK-8631 were taken and docked against AChE, BACE1. These fragments were further developed by fragment-based techniques. Quinolines, found to be active against AChE and $A\beta_{1-42}$ in our earlier study and piperazines, reported in many CNS active drugs were used as fragments. The later is also reported to improve the water solubility of small synthetic molecules without altering its BBB permeability. Linkers, substituted cyclohexane-1-amine or substituted benzyl-1-amine towards different amino acid residues were used to increase the approach of the tail group. The synthesized compounds were screened for *in-vitro* AChE, and BuChE inhibition, BACE1 inhibition, neuroprotection on MC65 cell lines, PAMPA assay, and pharmacokinetic studies on male Wistar rats.

The summary and conclusions of the study are included in chapter 8, which is followed by appendix and references.