Design and Development of Multi-target Directed 1,2,3-triazole-dimethylaminoacryloyl-chromenone Derivatives with Potential use in Alzheimer's Disease

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Abstract

To discover multifunctional agents for the treatment of Alzheimer’s disease (AD), a new series of 1,2,3-triazole-chromenone derivatives were designed and synthesized based on the multi target-directed ligands approach. The in vitro biological activities were evaluated including acetylcholinesterase (AChE), butyrylcholinesterase (BuChE), and A\textsubscript{β}\textsubscript{1–42} aggregation inhibition as well as neuroprotective effects and metal-chelating properties. The results indicated highly selective BuChE inhibitory activity with IC\textsubscript{50} values of 21.71 µM for compound 10 h as the most potent compound. Besides, compound 10 h could inhibit self-induced A\textsubscript{β}\textsubscript{1–42} aggregation and AChE-induced A\textsubscript{β} aggregation with 32.6% and 29.4% inhibition value, respectively. A Lineweaver–Burk plot and molecular modeling study also showed that compound 10 h targeted both the catalytic active site (CAS) and peripheral anionic site (PAS) of BuChE. It should be noted that compound 10 h was potent as a selective Cu\textsuperscript{2+} chelator. Thus, the designed scaffold could be considered as multifunctional agents for AD drug discovery developments.

Introduction

Dementia is one of the noteworthy problems in public health management as over 80% of dementia cases are suffering from Alzheimer’s disease (AD). Currently, available therapies provide temporary symptomatic relief but do not target the distractive neuropathology. Therefore, a new treatment to delay or halt disease progression has remained an urgent medical need.

The pathophysiological processes of AD remain still unclear to this day. However, alongside of its complexity, several neurodegenerative process during AD could be identified including (I) aggregation of insoluble β-amyloid (Aβ) plaques mostly trigger from sequential cleavage of amyloid precursor protein (APP) by the aspartyl protease β-site APP cleaving enzyme-1 (BACE1) and γ-secretase, (II) neurofibrillary tangles (NFTs) form through hyperphosphorylation of tau proteins, (III) biometals dysfunction, and (IV) oxidative stress which in return results in synapse loss and death of neuronal cells in the brain. Different hallmarks also have been recognized including the loss of cholinergic neurons, reduction of the neurotransmitter acetylcholine and increased expression of inflammatory mediators [1–3].

Based on the approved theory for AD, the loss of cholinergic neurons results in the reduction of the neurotransmitter acetylcholine (ACh). As a result, inhibition of the AChE raises the level of ACh and improves cognitive performance at the early stage of AD. The critical point is that AChE level decreases with the progression of AD, subsequently, AChE inhibition seems to be ineffective during the progression of AD [4, 5]. Interestingly, the level of the butyrylcholinesterase (BuChE) remains unchanged or even increases at the late stage of disease [6]. BuChE can hydrolyze ACh and, thereby, compensate the reduction of AChE activity [7]. An experiment with AChE knockout mice supported this hypothesis [8]. Results from further studies were in accordance with the role of BuChE in AD brains and showed a positive correlation between selective BuChE inhibition and improved cognitive performance and memory [9, 10].
Moreover, produced Aβ peptides can aggregate into Aβ plaques which initiate pathogenic cascade and lead to neuronal loss and dementia. Inhibition of the accumulation of Aβ peptide in the brain could be another therapeutic strategy against the development of AD \cite{1}. Metal chelatory potential of compounds has been also demonstrated to exert beneficial effects \textit{via} decreasing the plaque aggregation \cite{11, 12}.

**Results And Discussion**

**2.1 Design**

Because of multifactorial and sporadic nature of AD, the modern approach “multi-target – one disease” could be effective to develop an effective drug to simultaneously acting at different targets. Selective BuChE inhibitors could be a promising target for the treatment of AD at the moderate and advanced stages of the disease \cite{13}. Closer looks at X-ray crystallography of hBuChE depicted that it usually tolerates bigger scaffolds than hAChE as its active site of hBuChE is approximately 200 Å larger than hAChE. Analysis of the top potent hBuChE revealed that N-aromatic ring group is necessary for interactions with ChE active site (Fig. 1, A)\cite{14}; however, as appeared in compounds B and C increasing bulkiness and length of drug candidate could increase the selectivity of BuChE over hAChE. Besides, this part could also exhibit metal-chelating potential \cite{15, 16}. Moreover, introducing a relatively spacious dimethylamino propenone entity into our system would be a good strategy to increase selectivity toward BuChE.

In the case of anti-Aβ plaques aggregation, it is important to keep the potential moiety in the structure to inhibit the aggregation of the peptide.

Coumarin structures as active natural compounds may simultaneously possess anti-oxidative \cite{1, 17}, neuroprotective \cite{18} anti-ChE \cite{14} and anti-Aβ aggregative properties \cite{19, 20}. Coumarins pharmacophore owing to the presence of polar elements in the structure (Fig. 1, compound D and E) might help to inhibit amyloid fibril formation through interaction with the polar surface of Aβ \cite{21–23}. Hence, coumarins could serve as a rational framework for the prevention of Aβ\textsubscript{1-42} aggregation \cite{24}. In addition to the inhibition of the BuChE and Aβ plaques aggregation, an inhibitor that can tackle toxicity of AB peptide, ROS and RNS could be effective for a longer period of AD progression. Recently, iminochromene was characterized as potent neuroprotective agents. The iminochromene groups of compound F were bioisosterically replaced with chromenone moieties to evaluate the possible neuroprotectivity \cite{25}.

Hence, in the present work, a molecular hybridization and bioisosteric replacement approach were used to design multi-target agents with BuChE and Aβ aggregation inhibition, neuroprotective and metal chelating properties.

**2.2 Chemistry**

Synthesis of the tilted compounds 10 was conducted according to the steps shown in Scheme 1.
Desired starting material, 2-hydroxy-4-(prop-2-yn-1-yloxy)benzaldehyde (3) was exactly prepared according to the literature [26]. Then, the reaction of compound 3 and excess amounts of ethyl acetoacetate (4) in ethanol at room temperature overnight afforded 3-acetyl-7-(prop-2-yn-1-yloxy)-2H-chromen-2-one (5). Reaction of compound 5 and dimethylformamid-dimethylacetal (DMA-DMF) in 1,4-dioxane under reflux conditions for 6 h led to the formation of (E)-3-(3-(dimethylamino)acryloyl)-7-(prop-2-yn-1-yloxy)-2H-chromen-2-one (7). Finally, click reaction [27] of compound 7 and is suite prepared azides 9 in the presence of trimethylamine and CuSO₄·5H₂O in the mixture of water and tert-butyl alcohol at room temperature for 24 h gave the corresponding products 10a-m.

### 2.3 AChE And BuChE Inhibitory Activity

*In vitro* anti-AChE and anti-BuChE activity of synthesized compounds, 10a-m were performed based on the modified Ellman’s method [28] comparing with donepezil as the reference drug.

Compounds were initially screened *in vitro* against AChE, and none of them exhibited inhibitory properties against the AChE enzymes. Interestingly, all the 1,2,3-triazole-chromenone derivatives showed remarkable selectivity and potency towards BuChE, which exerted a more prominent role at later stages of the disease [29]. As can be seen in
Table 1, the inhibitory activity directly depended on the electronic property of substituents and their positions on the benzyl moiety connected to 1,2,3-triazole ring.

Results showed that compound 10 h possessing 3,4-diF on the aryl ring induced the best BuChEI activity ($IC_{50} = 21.71 \mu M$); however, the elimination of 3-F of the substituents completely changed the activity in such a manner that compound 10 g did not show any potency. *meta*-Fluorinated derivative 10f was found to be a moderate inhibitor as calculated $IC_{50}$ was 59.58 µM and *para*-fluorinated derivative 10 g showed no inhibitory activity toward BuChE ($IC_{50} > 100 \mu M$).

Considering the inhibitory activity of other halogenated derivatives 10i-m depicted that chlorinated compounds 10i and 10j showed no activity ($IC_{50} > 100 \mu M$). In the case of brominated derivatives 10 k-m, compounds 10 l possessing Br at 3- position of aryl ring showed moderate activity with $IC_{50} = 65.96 \mu M$. 

![Structural diagram](image)
Table 1
Anti-cholinesterase activity of 1,2,3-triazole-dimethylaminoacryloyl-chromenone 10a-m.a

| Entry | Ar        | Product 10 | AChEi IC₅₀ (µM) | BuChEi IC₅₀ (µM) |
|-------|-----------|------------|-----------------|------------------|
| 1     | C₆H₅      | 10a        | > 100           | 34.41 ± 0.23     |
| 2     | 2-Me-C₆H₄ | 10b        | > 100           | 35.73 ± 0.21     |
| 3     | 4-Me-C₆H₄ | 10c        | > 100           | > 100            |
| 4     | 3-MeO-C₆H₄| 10d        | > 100           | 23.44 ± 0.07     |
| 5     | 2-F-C₆H₄  | 10e        | > 100           | > 100            |
| 6     | 3-F-C₆H₄  | 10f        | > 100           | 59.58 ± 0.05     |
| 7     | 4-F-C₆H₄  | 10g        | > 100           | > 100            |
| 8     | 3,4-diF-C₆H₃| 10 h       | > 100           | 21.71 ± 0.57     |
|   | Substituent   | Compound | IC<sub>50</sub> (µM) | IC<sub>50</sub> (µM) |
|---|--------------|----------|-----------------------|-----------------------|
| 9 | 2-Cl-C<sub>6</sub>H<sub>4</sub> | 10i      | >100                  | >100                  |
| 10| 4-Cl-C<sub>6</sub>H<sub>4</sub> | 10j      | >100                  | >100                  |
| 11| 2-Br-C<sub>6</sub>H<sub>4</sub> | 10k      | >100                  | >100                  |
| 12| 3-Br-C<sub>6</sub>H<sub>4</sub> | 10l      | >100                  | 65.96 ± 0.004         |
| 13| 4-Br-C<sub>6</sub>H<sub>4</sub> | 10m      | >100                  | >100                  |
| 14| donepezil    |          | 0.079 ± 0.002         | 5.19 ± 0.38           |

*a Data are expressed as mean ± SD (three independent experiments).

In the case of the electron-donating substituent (Me), relatively good and selective results were obtained. The presence of methyl group at 2- position of aryl moiety (compound 10b) led to relatively good anti-BuChE activity (IC<sub>50</sub> = 35.73 µM); however, the presence of the same group at 4- position of compound 10c diminished inhibitory activity completely (IC<sub>50</sub> > 100 µM). It should be noted that the presence of strong electron-donating substituent (MeO) in compound 10d led to a higher activity than compounds 10b and 10c possessing methyl group (IC<sub>50</sub> = 23.44 µM).
Finally, the absence of substituent on the aryl ring (compound 10a) also depicted relatively good activity ($\text{IC}_{50} = 34.41 \, \mu\text{M}$).

The \textit{in vitro} anti-cholinesterase assay showed that the unsubstituted benzyl derivative (10a) along with the \textit{meta}-substituted analogues (10d, 10f, and 10 l) had significant anti-BuChE activity. Adding extra small-size halogen atoms such as fluorine (compound 10 h) resulted in the most potent activity with an $\text{IC}_{50}$ value of 21.71 \, \mu\text{M}.

2.4 Kinetic Study Of Buche Inhibition

The kinetic study was performed to investigate the mechanism of inhibition by compound 10 h against BuChE. Graphical analysis of the reciprocal Lineweaver–Burk plot of compound 10 h described a mixed-type inhibition pattern (Fig. 2, A.) in which compound 10 h may bound to the BuChE whether it already bound to the substrate. In addition, the $K_i$ value was calculated using the secondary plot as 28.2 \, \mu\text{M} (Fig. 2, B.).

2.5 Inhibition of AChE-induced and self-induced Aβ aggregation

Aβ peptide is the major constituent of senile plaques in the brains of patients with AD. In this respect, the effect of the most potent compound 10 h was assessed for the inhibition against Aβ$_{1-42}$ aggregations and AChE-induced Aβ$_{1-40}$ peptide aggregation using the Thioflavin T (ThT) assay. Comparing with donepezil and tacrine as the reference compounds, 10 h was more potent than both controls in inhibiting Aβ$_{1-42}$ self-aggregation, as depicted 32.6% values at 10 \, \mu\text{M} (Table 2.). Furthermore, compound 10 h inhibited AChE-induced Aβ aggregation by 29.4% at 100 \, \mu\text{M}.
Table 2
Inhibitory activities of compounds 10 h against Aβ1–42 aggregation

| Samples       | % Inhibition self-induced Aβ1–42 aggregation | % Inhibition AChE-induced Aβ aggregation |
|---------------|---------------------------------------------|-----------------------------------------|
| 10 h          | 32.6 ± 2.0                                  | 29.4 ± 1.5                               |
| Tacrine       | 7.6 ± 1.4                                   | 6.7 ± 0.9                                |
| Donepezil     | 18.1 ± 1.4 (10 µM)                          | 25.2 ± 1.7 (100 µM)                      |

a Values are expressed as means ± SEM of three experiments.

b Inhibition of self-induced Aβ(1–42) aggregation (25 mM) produced by the tested compound at 10 µM concentration.

c Co-aggregation inhibition of Aβ(1–40) and AChE (2 µM, ratio 100:1) by the tested compound at 100 µM.

2.6 Neuroprotective Studies On Pc12 Cell Line

Compound 10 h was selected to study the neuroprotective ability using PC12 cell injury induced by Aβ25–35 by MTT assay. This compound depicted no neuroprotective effect on Aβ-induced PC12 cells up to 50 µM. It can be understood that bioisosterically replacement of iminochromene moiety (compound F, Fig. 1.) with chromenone did not induce desired neuroprotectivity.

2.7 Metal Chelating

Compound 10 h was tested for its metal chelating ability towards Fe²⁺, Cu²⁺, and Zn²⁺ ions (Fig. 3.) The UV spectrum of methanolic solution (20 µM) of that compound showed two characteristic absorption peaks at 309.9 and 386.7 nm. After the interaction of compound 10 h with the abovementioned ions for 30 min, red and blue shifts observed in the spectra confirmed desired interactions of that compound with biometals. Interaction of compound 10 h with Zn²⁺ ions demonstrated two absorption peaks at 299.2 and 388.7 nm which demonstrated red and blue shifts, respectively. Similar changes were observed in the case of Fe²⁺ ions and those absorptions were observed at 303.5 and 390.9 nm. Also, another absorption peak was obtained at 209.6 nm. When compound 10 h was treated with Cu²⁺ ions, blue shifts were observed at 303.5 and 382.4 nm and a peak at 205.3 was also observed.

The stoichiometry of complex 10 h-Cu²⁺ was also studied (Fig. 4.). The concentration of the test compound 10 h was 20 µM and the final concentration of Cu²⁺ ranged from 0–20 µM with 4 µM intervals at 205.3 nm. The plot was obtained by the corresponding absorption against the mole fraction of Cu²⁺ to
ligand 10 h. According to the plot, the ratio 1:1 complexation ration of 10 h-Cu$^{2+}$ can be seen at the fracture point of the plot with the mole fraction of 0.6.

### 2.8 Docking Study Of BuChE

The volume of the BuChE active site gorge is considerably higher than the one found in AChE, so BuChE can accommodate bulkier inhibitors, and this may constitute the basis for the selectivity of these derivatives. An overlay of the best pose for 10 h with BuChE was depicted in Fig. 5, chromenenone core demonstrated π-π stacking interactions with Trp82, Gly117, and Phe329.

Carbonyl group of the mentioned ring allowed the oxygen to form a hydrogen bond with the hydroxyl on Ser198 of catalytic triads at 2.27 Å while 1,2,3-triazole ring formed a second hydrogen bond to Trp82 of anionic subside. The nitrogen of the dimethylamino acryloyl chromenone interacted with Pro285 via van der Waals interactions. Further π-π stacking interaction was constructed between 1,2,3-triazole moiety and Phe329 and Met437 with the PAS residue. The para-fluoro benzyl ring also showed π – aryl interaction with Ala328.

### 2.9 In silico ADME evaluation

The synthesized compounds were further assessed in terms of physiochemical parameters and pharmacokinetic properties using http://lmmd.ecust.edu.cn/admetsar2/ and http://preadmet.bmdrc.kr).
Table 3
Calculated molecular profile for synthesized compounds.

| Descriptor | Compound | Mw   | ClogP | H-Bond Acceptor | H-Bond Donor | BBB    | Human Intestinal Absorption | Caco-2 permeability |
|------------|----------|------|-------|-----------------|--------------|--------|----------------------------|---------------------|
| 10a        |          | 430.46 | 3.27  | 8               | 0            | +      | 0.9757                     | −0.8029             |
| 10b        |          | 444.49 | 3.58  | 8               | 0            | +      | 0.9754                     | −0.7985             |
| 10c        |          | 444.49 | 3.58  | 8               | 0            | +      | 0.9757                     | −0.7908             |
| 10d        |          | 460.49 | 3.28  | 9               | 0            | +      | 0.9757                     | −0.7587             |
| 10e        |          | 448.45 | 3.41  | 8               | 0            | +      | 0.9757                     | −0.8157             |
| 10f        |          | 448.45 | 3.41  | 8               | 0            | +      | 0.9757                     | −0.7970             |
| 10g        |          | 448.45 | 3.41  | 8               | 0            | +      | 0.9757                     | −0.8152             |
| 10h        |          | 466.44 | 3.55  | 8               | 0            | +      | 0.9757                     | −0.8211             |
| 10i        |          | 464.91 | 3.92  | 8               | 0            | +      | 0.9746                     | −0.8289             |
| 10j        |          | 464.91 | 3.92  | 8               | 0            | +      | 0.9746                     | −0.8211             |
| 10k        |          | 509.36 | 4.03  | 8               | 0            | +      | 0.9751                     | −0.8299             |
| 10l        |          | 509.36 | 4.03  | 8               | 0            | +      | 0.9751                     | −0.8115             |
| 10m        |          | 509.36 | 4.03  | 8               | 0            | +      | 0.9751                     | −0.8306             |

As shown in Table 3, most of compounds showed drug-like characteristics based on Lipinski’s rule of five (MW < 500, C-log P < 5, HB donor ≤ 5, HB acceptor ≤ 10). Our results indicated that lipophilicity and solubility of the derivatives were drug-like. Furthermore molecular weight, C-logP, and blood-brain barrier were well within the standard ranges.

Conclusion

In summary, a series of 1,2,3-triazole-dimethylaminoacyloyl-chromenone derivatives were designed and synthesized as multifunctional anti-Alzheimer’s agents. All the target compounds were synthesized and
screened as AChE/BuChE inhibition. The most active compound in this series was further evaluated by multiple biological activities including, \( \text{A}\beta_{1-42} \) aggregation inhibition, metal-chelating properties and neuroprotective effects against \( \text{A}\beta_{25-35} \)-induced PC12 cell injury. Our results showed that these compounds had a high inhibitory potency and selectivity toward BuChE with an IC\(_{50}\) value of 21.71 \( \mu \text{M} \) for 10 h as the most potent BuChE inhibitor. The inhibition kinetic analysis revealed a mixed-type inhibition pattern for this compound. The molecular modeling study of BuChE for 10 h indicated that the mentioned compound 10 h bound to both CAS and PAS of the BuChE. Moreover, this compound had a significant and anti-\( \text{A}\beta \) aggregation capacity. Compound 10 h served as a selective metal chelator by chelating Cu\(^{2+}\).

Therefore, the results indicated that this hybridization approach could be successful strategy for further developments of potential multifunctional candidates against AD.

**Materials And Methods**

**4.1 Chemistry**

Melting points of synthesized compounds were determined on a Kofler hot stage apparatus. \(^1\text{H}\) and \(^{13}\text{C}\) NMR spectra were determined on a Varian FT-500, using TMS as an internal standard. IR spectra were recorded using KBr disks on a Bruker Tensor 27 FTIR spectrophotometer. Elemental analysis was carried out with an Elemental Analyzer system GmbH VarioEL CHN mode.

**Synthesis of 2-hydroxy-4-(prop-2-yn-1-yloxy)benzaldehyde (3)**

Compound 3 was prepared from the reaction of 2,4-dihydroxybenzaldehyde 1 and propargyl bromide 2 in the presence of potassium carbonate (\( \text{K}_2\text{CO}_3 \)) and potassium iodide (KI) in acetone at 50 °C, according to the literature [26].

**Synthesis of 3-acetyl-7-(prop-2-yn-1-yloxy)-2\( \text{H}\)-chromen-2-one (5)**

A few drops of piperidine was added to the mixture of compound 3 (1 mmol) and ethyl acetoacetate (2.5 mmol) 4 in ethanol (10 mL) and it was stirred overnight at room temperature to obtain yellow precipitates. After completion of reaction (checked by TLC), they were filtered off and used for the next step with no further purification.

**Synthesis of (E)-3-(3-(dimethylamino)acryloyl)-7-(prop-2-yn-1-yloxy)-2\( \text{H}\)-chromen-2-one (7)**

The mixture of compound 5 (1 mmol) and DMA-DMF 6 (2 mmol) in 1,4-dioxane (10 mL) was heated at reflux for 6 h. Then, the solvent was evaporated under vacuum and the residue was purified using plate chromatography with ethyl acetate as eluent.

**Synthesis of 1,2,3-triazole-dimethylaminoacryloyl-chromenone hybrids 10a-m**
The final step was performed by the click reaction of compound 7 and is suite prepared azides 9. For this purpose, a solution of benzyl chloride/bromide derivative 8 (1.1 mmol), sodium azide (0.06 g, 0.9 mmol), and trimethylamine (0.13 g, 1.3 mmol) in water (4 mL) and tert-butyl alcohol (4 mL) was stirred at room temperature for 30 min. Then, compound 7 (0.5 mmol) and CuSO$_4$.5H$_2$O (7 mol%) were added to the mixture and it was continued for 24 h. Upon completion of the reaction checked TLC), the mixture was diluted with water, extracted with chloroform, and dried over anhydrous Na$_2$SO$_4$. After evaporation of solvent, the residue was recrystallized from ethyl acetate and petroleum ether to give pure product 10. In the case of some compounds, they were purified using plate chromatography with ethyl acetate as eluent.

(E)-7-((1-Benzyl-1H-1,2,3-triazol-4-yl)methoxy)-3-(3-(dimethylamino)acryloyl)-2H-chromen-2-one (10a)

Yield: 54%; M.p. 186-188 °C. IR (KBr): 2920, 2853, 1715, 1640, 1597, 1558 cm$^{-1}$. $^1$H NMR (CDCl$_3$-d$_6$, 500 MHz): $\delta$ = 8.55 (s, 1H, triazole), 7.91 (d, $J$ = 12.5 Hz, 1H, CH), 7.60 (s, 1H, H4), 7.50 (d, $J$ = 8.5 Hz, H5), 7.38-7.29 (m, 5H, H2′, H3′, H4′, H5′, H6′), 6.92-6.90 (m, 2H, H6, H8), 6.33 (d, $J$ = 12.5 Hz, 1H, CH), 5.55 (s, 2H, CH$_2$), 5.24 (s, 2H, CH$_2$), 3.17 (s, 3H, CH$_3$), 2.96 (s, 3H, CH$_3$) ppm. $^{13}$C NMR (CDCl$_3$-d$_6$, 125 MHz): $\delta$ = 182.4, 171.7, 162.9, 156.2, 151.4, 145.6, 144.0, 134.7, 133.3, 131.1, 129.8, 128.9, 128.2, 122.6, 121.5, 113.3, 101.2, 92.2, 63.4, 57.2, 47.6, 38.1 ppm. Anal. calcd. for C$_{24}$H$_{22}$N$_4$O$_4$: C, 66.97; H, 5.15; N, 13.02. Found: C, 66.71; H, 5.38; N, 12.86.

(E)-3-(3-(Dimethylamino)acryloyl)-7-((1-(2-methylbenzyl)-1H-1,2,3-triazol-4-yl)methoxy)-2H-chromen-2-one (10b)

Yield: 59%; M.p. 203-205 °C. IR (KBr): 2924, 2855, 1712, 1640, 1596, 1558 cm$^{-1}$. $^1$H NMR (CDCl$_3$-d$_6$, 500 MHz): $\delta$ = 8.58 (s, 1H, triazole), 7.92 (d, $J$ = 12.3 Hz, 1H, CH), 7.52 (d, 1H, $J$ = 8.3 Hz, H5), 7.46 (s, 1H, H4), 7.32-7.18 (m, 4H, H3′, H4′, H5′, H6′), 6.93-6.91 (m, 2H, H6, H8), 6.35 (d, $J$ = 12.3 Hz, 1H, CH), 5.57 (s, 2H, CH$_2$), 5.24 (s, 2H, CH$_2$), 3.18 (s, 3H, CH$_3$), 2.98 (s, 3H, CH$_3$), 2.29 (s, 3H, CH$_3$) ppm. $^{13}$C NMR (CDCl$_3$-d$_6$, 125 MHz): $\delta$ = 182.4, 171.7, 162.9, 156.2, 151.4, 145.6, 144.0, 134.7, 133.3, 131.1, 129.8, 128.9, 128.2, 122.6, 121.5, 113.3, 101.2, 92.2, 63.4, 57.2, 47.6, 38.1 ppm. Anal. calcd. for C$_{25}$H$_{24}$N$_4$O$_4$: C, 67.55; H, 5.44; N, 12.60. Found: C, 67.31; H, 5.70; N, 12.44.

(E)-3-(3-(Dimethylamino)acryloyl)-7-((1-(4-methylbenzyl)-1H-1,2,3-triazol-4-yl)methoxy)-2H-chromen-2-one (10c)

Yield: 55%; M.p. 199-201 °C. IR (KBr): 2923, 2855, 1715, 1640, 1596, 1558 cm$^{-1}$. $^1$H NMR (CDCl$_3$-d$_6$, 500 MHz): $\delta$ = 8.57 (s, 1H, triazole), 7.92 (d, $J$ = 12.3 Hz, 1H, CH), 7.55 (s, 1H, H4), 7.51 (d, 1H, $J$ = 8.3 Hz, H5), 7.19-7.17 (m, 4H, H2′, H3′, H5′, H6′), 6.94-6.91 (m, 2H, H6, H8), 6.34 (d, $J$ = 12.3 Hz, 1H, CH), 5.51 (s, 2H, CH$_2$), 5.24 (s, 2H, CH$_2$), 3.17 (s, 3H, CH$_3$), 2.97 (s, 3H, CH$_3$), 2.36 (s, 3H, CH$_3$) ppm. $^{13}$C NMR (CDCl$_3$-d$_6$, 125 MHz): $\delta$ = 182.5, 162.4, 160.2, 156.6, 1549, 145.9, 143.1, 138.9, 131.2, 130.7, 129.8, 128.2, 123.3, 122.8,
113.3, 133.2, 101.2, 95.2, 62.4, 54.6, 45.6, 37.6, 21.2 ppm. Anal. calcd. for C_{25}H_{24}N_{4}O_{4}: C, 67.55; H, 5.44; N, 12.60. Found: C, 67.40; H, 5.26; N, 12.71.

**(E)-3-(3-(Dimethylamino)acryloyl)-7-((1-(3-methoxybenzyl)-1H-1,2,3-triazol-4-yl)methoxy)-2H-chromen-2-one (10d)**

Yield: 52%; M.p. 209-211 °C. IR (KBr): 2923, 2855, 1712, 1640, 1595, 1557 cm⁻¹. ¹H NMR (CDCl₃-d₆, 500 MHz): δ = 8.57 (s, 1H, triazole), 7.92 (d, J = 12.4 Hz, 1H, CH), 7.59 (s, 1H, H₄), 7.52 (d, 1H, J = 8.4 Hz, H₅), 7.30 (t, J = 7.9 Hz, 1H, H₅'), 6.94-6.81 (m, 5H, H₆, H₈, H₂', H₄', H₆'), 6.35 (d, J = 12.4 Hz, 1H, CH), 5.52 (s, 2H, CH₂), 5.25 (s, 2H, CH₂), 3.79 (s, 3H, OCH₃), 3.17 (s, 3H, CH₃), 2.97 (s, 3H, CH₃) ppm. ¹³C NMR (CDCl₃-d₆, 125 MHz): δ = 182.4, 162.4, 160.2, 156.6, 154.9, 145.9, 144.0, 143.2, 135.7, 130.7, 130.3, 123.4, 122.9, 119.3, 114.3, 113.8, 133.3, 112.5, 101.2, 95.2, 62.4, 55.3, 54.3, 45.2, 37.6 ppm. Anal. calcd. for C_{25}H_{24}N_{4}O_{4}: C, 67.40; H, 5.26; N, 12.71.

**(E)-3-(3-(Dimethylamino)acryloyl)-7-((1-(2-fluorobenzyl)-1H-1,2,3-triazol-4-yl)methoxy)-2H-chromen-2-one (10e)**

Yield: 55%; M.p. 180-183 °C. IR (KBr): 2924, 2852, 1712, 1640, 1597, 1549 cm⁻¹. ¹H NMR (CDCl₃-d₆, 500 MHz): δ = 8.57 (s, 1H, triazole), 7.91 (d, J = 12.3 Hz, 1H, CH), 7.51 (d, 1H, J = 8.0 Hz, H₅), 7.39-7.38 (m, 1H, H₄'), 6.94-6.92 (m, 2H, H₅′, H₆'), 7.17-7.11 (m, 2H, H₅′, H₆′), 6.97 (d, 1H, J = 9.2 Hz, H₂′), 6.94-6.91 (m, 2H, H₆, H₈), 6.34 (d, J = 12.3 Hz, 1H, CH), 5.61 (s, 2H, CH₂), 5.25 (s, 2H, CH₂), 3.17 (s, 3H, CH₃), 2.97 (s, 3H, CH₃) ppm. ¹³C NMR (CDCl₃-d₆, 125 MHz): δ = 182.3, 162.4, 160.6 (d, J_C-F = 251.2 Hz), 160.1, 158.7, 156.6, 154.8, 145.9, 143.2, 131.1 (d, J_C-F = 8.1 Hz), 130.7 (d, J_C-F = 3.3 Hz), 124.9 (d, J_C-F = 3.4 Hz), 123.3, 121.3, 121.6 (d, J_C-F = 14.5 Hz), 115.9 (d, J_C-F = 21.03 Hz), 113.3, 113.2, 101.2, 95.2, 62.4, 47.9, 45.2, 37.5 ppm. Anal. calcd. for C_{24}H_{21}FN_{4}O_{4}: C, 64.36; H, 4.72; N, 12.49. Found: C, 64.36; H, 4.57; N, 12.55.

**(E)-3-(3-(Dimethylamino)acryloyl)-7-((1-(3-fluorobenzyl)-1H-1,2,3-triazol-4-yl)methoxy)-2H-chromen-2-one (10f)**

Yield: 60%; M.p. 183-185 °C. IR (KBr): 2922, 2850, 1713, 1640, 1597, 1557 cm⁻¹. ¹H NMR (CDCl₃-d₆, 500 MHz): δ = 8.57 (s, 1H, triazole), 7.91 (d, J = 12.3 Hz, 1H, CH), 7.51 (d, 1H, J = 8.0 Hz, H₅), 7.39-7.38 (m, 1H, H₄'), 7.30 (td, J = 7.6, 1.5 Hz, 1H, H₃'), 7.17-7.11 (m, 2H, H₅', H₆'), 6.94-6.92 (m, 2H, H₆, H₈), 6.34 (d, J = 12.4 Hz, 1H, CH), 5.61 (s, 2H, CH₂), 5.25 (s, 2H, CH₂), 3.17 (s, 3H, CH₃), 2.97 (s, 3H, CH₃) ppm. ¹³C NMR (CDCl₃-d₆, 125 MHz): δ = 180.0, 162.3, 160.7 (d, J_C-F = 262.2 Hz), 160.1, 156.7, 154.9, 145.8, 143.5, 136.6 (d, J_C-F = 19.4 Hz), 130.8 (d, J_C-F = 8.1 Hz), 130.7, 123.6, 123.3, 122.9, 115.9 (d, J_C-F = 20.9 Hz), 115.1 (d, J_C-F = 22.0 Hz), 113.3, 113.2, 101.2, 95.2, 62.4, 53.7, 45.0, 37.6 ppm. Anal. calcd. for C_{24}H_{21}FN_{4}O_{4}: C, 64.28; H, 4.72; N, 12.49. Found: C, 64.47; H, 4.84; N, 12.60.
(E)-3-(3-(Dimethylamino)acryloyl)-7-((1-(4-fluorobenzyl)-1H-1,2,3-triazol-4-yl)methoxy)-2H-chromen-2-one (10g)

Yield: 62%; M.p. 183-185 °C. IR (KBr): 2923, 2855, 1714, 1640, 1598 cm⁻¹. ¹H NMR (CDCl₃-d₆, 500 MHz): δ = 8.56 (s, 1H, triazole), 7.92 (d, J = 12.4 Hz, 1H, CH), 7.58 (s, 1H, H₄), 7.51 (d, 1H, J = 8.4 Hz, H₅), 7.29 (dd, J = 8.7, 5.1 Hz, 2H, H₂', H₆'), 7.07 (t, J = 8.7 Hz, 2H, H₃', H₅'), 6.93-6.91 (m, 2H, H₂, H₆, H₈), 6.33 (d, J = 12.4 Hz, 1H, CH), 5.52 (s, 2H, CH₂), 5.24 (s, 2H, CH₂), 3.16 (s, 3H, CH₃), 2.96 (s, 3H, CH₃) ppm. ¹³C NMR (CDCl₃-d₆, 125 MHz): δ = 180.1, 162.4, 160.5 (d, J_C-F = 260.5 Hz), 160.1, 156.5, 154.8, 145.1, 143.4, 134.5 (d, J_C-F = 19.5 Hz), 130.7, 130.1, 123.3, 122.8, 116.2 (d, J_C-F = 21.0 Hz), 113.3, 113.2, 101.2, 95.1, 62.1, 53.5, 45.0, 37.5 ppm. Anal. calcd. for C_{24}H_{21}FN_{4}O_{4}: C, 64.28; H, 4.72; N, 12.49. Found: C, 64.44; H, 4.60; N, 12.27.

(E)-7-((1-(3,4-Difluorobenzyl)-1H-1,2,3-triazol-4-yl)methoxy)-3-(3-(dimethylamino)acryloyl)-2H-chromen-2-one (10h)

Yield: 59%; M.p. 188-190 °C. IR (KBr): 2922, 2850, 1718, 1640, 1594 cm⁻¹. ¹H NMR (CDCl₃-d₆, 500 MHz): δ = 8.57 (s, 1H, triazole), 7.91 (d, J = 12.3 Hz, 1H, CH), 7.63 (s, 1H, H₄), 7.52 (d, 1H, J = 8.4 Hz, H₅), 7.21-7.11 (m, 2H, H₅', H₆'), 7.06-7.04 (m, 1H, H₂'), 6.93-6.91 (m, 2H, H₆, H₈), 6.33 (d, J = 12.3 Hz, 1H, CH), 5.52 (s, 2H, CH₂), 5.27 (s, 2H, CH₂), 3.18 (s, 3H, CH₃), 2.97 (s, 3H, CH₃) ppm. ¹³C NMR (CDCl₃-d₆, 125 MHz): δ = 182.3, 160.1, 156.6, 154.9, 150.5 (d, J_C-F = 243.1 Hz), 149.6, (d, J_C-F = 240.0 Hz), 145.8, 144.0, 143.6, 132.8, 131.2, 130.7, 124.3, 123.4, 122.9, 118.1 (d, J_C-F = 17.2), 117.3 (d, J_C-F = 17.6), 113.3, 101.2, 95.2, 62.4, 53.2, 45.2, 37.6 ppm. Anal. calcd. for C_{24}H_{20}F₂N₄O₄: C, 61.80; H, 4.32; N, 12.01. Found: C, 61.63; H, 4.17; N, 11.84.

(E)-7-((1-(2-Chlorobenzyl)-1H-1,2,3-triazol-4-yl)methoxy)-3-(3-(dimethylamino)acryloyl)-2H-chromen-2-one (10i)

Yield: 58%; M.p. 178-180 °C. IR (KBr): 2923, 2852, 1714, 1640, 1597 cm⁻¹. ¹H NMR (CDCl₃-d₆, 500 MHz): δ = 8.60 (s, 1H, triazole), 7.91 (d, J = 11.0 Hz, 1H, CH), 7.68 (s, 1H, H₄), 7.51 (d, 1H, J = 8.1 Hz, H₅), 7.43 (d, 1H, J = 7.9 Hz, H₃'), 7.32-7.24 (m, 3H, H₄', H₅', H₆'), 6.93-6.87 (m, 2H, H₆, H₈), 6.33 (d, J = 11.0 Hz, 1H, CH), 5.69 (s, 2H, CH₂), 5.29 (s, 2H, CH₂), 3.16 (s, 3H, CH₃), 2.96 (s, 3H, CH₃) ppm. ¹³C NMR (CDCl₃-d₆, 125 MHz): δ = 182.7, 171.5, 163.8, 158.5, 156.3, 151.7, 144.5, 133.6, 131.7, 130.7, 130.5, 130.4, 130.0, 127.7, 123.4, 120.1, 113.3, 112.8, 100.6, 96.6, 61.1, 52.3, 46.4, 40.4 ppm. Anal. calcd. for C_{24}H_{21}ClN_{4}O_{4}: C, 62.00; H, 4.55; N, 12.05. Found: C, 61.81; H, 4.38; N, 11.90.

(E)-7-((1-(4-Chlorobenzyl)-1H-1,2,3-triazol-4-yl)methoxy)-3-(3-(dimethylamino)acryloyl)-2H-chromen-2-one (10j)

Yield: 61%; M.p. 178-180 °C. IR (KBr): 2891, 2850, 1715, 1640, 1597, 1558 cm⁻¹. ¹H NMR (CDCl₃-d₆, 500 MHz): δ = 8.54 (s, 1H, triazole), 7.90 (d, J = 12.4 Hz, 1H, CH), 7.61 (s, 1H, H₄), 7.50 (d, J = 8.5 Hz, 1H, H₅), 7.34 (d, J = 8.1 Hz, 2H, H₃', H₅'), 7.22 (d, J = 8.1 Hz, 2H, H₂', H₆').
6.92-6.89 (m, 2H, H6, H8), 6.34 (d, J = 12.4 Hz, 1H, CH), 5.52 (s, 2H, CH2), 5.19 (s, 2H, CH2), 3.11 (s, 3H, CH3), 2.95 (s, 3H, CH3) ppm. 13C NMR (CDCl3-d6, 125 MHz): δ = 182.9, 162.4, 160.1, 156.5, 154.9, 145.8, 144.2, 143.4, 134.9, 130.7, 129.5, 129.4, 123.3, 122.9, 113.3, 133.2, 101.2, 95.2, 62.4, 53.5, 45.2, 37.6 ppm. Anal. calcd. for C24H21ClN4O4: C, 62.00; H, 4.55; N, 12.05. Found: C, 62.11; H, 4.40; N, 12.28.

(E)-7-((1-(2-Bromobenzyl)-1H-1,2,3-triazol-4-yl)methoxy)-3-(3-(dimethylamino)acryloyl)-2H-chromen-2-one (10k)

Yield: 58%; M.p. 202-205 °C. IR (KBr): 2924, 2850, 1709, 1641, 1598, 1559 cm⁻¹. 1H NMR (CDCl3-d6, 500 MHz): δ = 8.57 (s, 1H, triazole), 7.92 (d, J = 12.3 Hz, 1H, CH), 7.70 (s, 1H, H4), 7.63 (d, 1H, J = 7.5 Hz, H3'), 7.52 (d, 1H, J = 8.3 Hz, H5), 7.33 (t, 1H, J = 7.5 Hz, H5'), 7.25-7.21 (m, 2H, H4', H6'), 6.95-6.93 (m, 2H, H6, H8), 6.34 (d, J = 12.3 Hz, 1H, CH), 5.69 (s, 2H, CH2), 5.27 (s, 2H, CH2), 3.21 (s, 3H, CH3), 2.97 (s, 3H, CH3) ppm. 13C NMR (CDCl3-d6, 125 MHz): δ = 182.5, 162.4, 160.1, 156.6, 154.9, 145.9, 143.1, 133.8, 133.4, 133.3, 130.7, 130.6, 130.5, 128.3, 123.6, 123.3, 113.4, 113.3, 100.4, 95.2, 62.0, 54.0, 44.8, 37.2 ppm. Anal. calcd. for C24H21BrN4O4: C, 56.59; H, 4.16; N, 11.00. Found: C, 56.37; H, 4.30; N, 11.21.

(E)-7-((1-(3-Bromobenzyl)-1H-1,2,3-triazol-4-yl)methoxy)-3-(3-(dimethylamino)acryloyl)-2H-chromen-2-one (10l)

Yield: 64%; M.p. 218-220 °C. IR (KBr): 2925, 2856, 1710, 1640, 1595 cm⁻¹. 1H NMR (CDCl3-d6, 500 MHz): δ = 8.59 (s, 1H, triazole), 7.93 (d, J = 12.4 Hz, 1H, CH), 7.61 (s, 1H, H4), 7.55-7.51 (m, 3H, H5, H2', H4'), 7.26-7.18 (m, 2H, H5', H6'), 6.95-6.93 (m, 2H, H6, H8), 6.35 (d, J = 12.4 Hz, 1H, CH), 5.51 (s, 2H, CH2), 5.28 (s, 2H, CH2), 3.19 (s, 3H, CH3), 2.98 (s, 3H, CH3) ppm. 13C NMR (CDCl3-d6, 125 MHz): δ = 180.1, 162.4, 160.1, 156.6, 154.9, 145.9, 143.1, 133.8, 133.4, 133.3, 130.7, 130.6, 130.5, 128.3, 123.6, 123.3, 113.4, 113.3, 100.4, 95.2, 62.0, 54.0, 44.8, 37.2 ppm. Anal. calcd. for C24H21BrN4O4: C, 56.59; H, 4.16; N, 11.00. Found: C, 56.31; H, 4.24; N, 11.18.

(E)-7-((1-(4-Bromobenzyl)-1H-1,2,3-triazol-4-yl)methoxy)-3-(3-(dimethylamino)acryloyl)-2H-chromen-2-one (10m)

Yield: 64%; M.p. 173-175 °C. IR (KBr): 2925, 2850, 1715, 1640, 1597 cm⁻¹. 1H NMR (CDCl3-d6, 500 MHz): δ = 8.57 (s, 1H, triazole), 7.91 (d, J = 12.3 Hz, 1H, CH), 7.60 (s, 1H, H4), 7.52-7.51 (m, 3H, H5, H3', H5'), 7.17 (d, J = 8.4 Hz, 2H, H2', H6'), 6.93-6.91 (m, 2H, H6, H8), 6.34 (d, J = 12.3 Hz, 1H, CH), 5.51 (s, 2H, CH2), 5.22 (s, 2H, CH2), 3.17 (s, 3H, CH3), 2.96 (s, 3H, CH3) ppm. 13C NMR (CDCl3-d6, 125 MHz): δ = 182.4, 162.9, 160.1, 156.6, 154.9, 145.9, 143.5, 136.4, 133.7, 132.1, 131.1, 130.9, 130.8, 126.6, 122.9, 122.2, 113.4, 113.3, 101.1, 95.0, 62.8, 53.6, 44.8, 38.8 ppm. Anal. calcd. for C24H21BrN4O4: C, 56.59; H, 4.16; N, 11.00. Found: C, 56.31; H, 4.24; N, 11.18.

4.2 Inhibitory activities against AChE and BuChE
All enzymes and reagent required for the assay was obtained from Aldrich. The in vitro anticholinesterase activity of all synthesized compounds 10a-m was assayed using modified Ellman's method using a 96-well plate reader (BioTek ELx808) according to the literature [28, 30]. Initially, the stock solutions of compounds 10 were prepared by dissolving of the test compound (1 mg) in DMSO (1 mL) and then, diluted solutions at final concentrations of 1, 10, 20, and 40 μg/mL were prepared using methanol. Each well contained 50 μL potassium phosphate buffer (KH₂PO₄/ K₂HPO₄, 0.1 M, pH 8), 25 μL sample solution, and 25 μL enzyme (final concentration 0.22 U/mL in buffer). Control experiments were also performed under the same conditions without enzyme. After incubation at room temperature for 15 min, 125 μL DTNB (3 mM in buffer) was added and characterization of enzymatic reaction was spectrometrically performed at 405 nm followed by the addition of substrate (ATCl 3 mM in water) after 5-10 min. The IC₅₀ values were determined graphically from inhibition curves (log inhibitor concentration vs. percent of inhibition). Also, the same method was also used for BChE inhibition assay.

4.3 Kinetic characterization of BuChE inhibitory activity

The kinetic study for the inhibition of BuChE by compound 10h was carried out according to the Ellman's method used for the inhibition assay using four different concentrations of inhibitor (0, 10.7, 42.9, and 85.8 μM). The Lineweaver–Burk reciprocal plot was provided by plotting 1/V against 1/[S] at variable concentrations of the substrate butyrylthiocholine (187.5, 750, 1500, 3000 μM). The inhibition constant Ki was achieved by the plot of slopes versus the corresponding concentrations of the compound 10h [31, 32].

4.4 Inhibition of Aβ₁-₄₂ aggregation and disaggregation of aggregated Aβ₁-₄₀ induced by AchE

Inhibition of Aβ1-42 self-aggregation measured by ThT fluorescence assay. The details of method were reported in our previous study [33]. To study Aβ₄₂ aggregation inhibition, a reported method, based on the fluorescence emission of ThT was followed. Briefly, the mixtures of Aβ₁₄₀ peptide (Bachem company, Switzerland) and AChE (Sigma, Electrophorus electricus), in presence or absence of the test inhibitor were incubated for 24 h at room temperature. The final concentrations of Aβ (dissolved in DMSO and diluted 0.215 M sodium phosphate buffer, pH 8), AChE (dissolved in 0.215 M sodium phosphate buffer, pH 8.0) and the tested compound are 200 μM, 2 μM and 100 μM respective. After co-incubation, 20 μL of the mixture solutions was diluted to a final volume of 2 mL with ThT (1.5 μM in 50 mM glycine-NaOH buffer, pH 8.5) was measured with multi-mode plate reader at the excitation and emission wavelength of λex = 450 nm and λem = 485 nm, respectively [33].

4.5 Neuroprotection assay against Aβ-induced damage
MTT reduction assay was used to evaluate the neuroprotective effect of 10h on neuronal PC12 cells damage induced by Aβ25-35. The cells were grown in monolayer culture on collagen-coated plates at 37 °C in a humidified atmosphere of 5% CO2. Neuronal PC12 cells were plated at a density of 5 × 10^5 cells/well on 96-well plates. The cells were pre-incubated with 10h for 3 h before human Aβ25–35 (final concentration of 5 μM) was added. After 24 h, 90 µl the medium was taken out and 20 µL of MTT (0.5 mg/ml dissolved in RPMI containing phenol red) was added and incubated for an additional 2 h at 37 °C. The absorbance (A570 nm) was measured using a Bio-Rad microplate reader (Model 680, Bio-Rad). The details referenced our previous work [12, 25].

4.6 Metal chelation studies

To study the metal chelating ability, the solutions of compound 10h and Fe2+, Cu2+, and Zn2+ ions (from FeSO4, CuCl2, and ZnCl2) were prepared in methanol. The mixture of compound 10h (1 mL) and the test ion solutions (1 mL) with the same final concentration of 20 µM in a 1 cm quartz cuvette was incubated at room temperature for 30 min. Then, the absorption spectra were recorded with wavelength ranging from 200–600 nm. The stoichiometry of complex 10h-Cu2+ was also studied using the molar ratio method [11, 34]. The concentration of compound 10h was 20 µM and the final concentration of Cu2+ ranged from 0-20 µM with 4 µM intervals at 205.3 nm. The plot was obtained by the corresponding absorption versus mole fraction of Cu2+ to ligand 10h.

4.7 Molecular docking

The molecular docking studies of the most potential ligand was performed on BuChE (PDB code: 4BDS) to observe the binding orientation and consensual binding interactions using AutoDock 4.2. The X-ray crystal structure of receptor was downloaded from the PDB database. All water and ligand molecules were removed from the structure, and the protein was prepared for docking. The co-crystallized ligand within the pdb structures was defined as a center of the binding site. All ligands were created using Chem3D Ultra software, and energy minimizations were done by the semiempirical MM^[35]. The compounds were docked into the active site of proteins using default parameters for each ligand with 100 runs and 27,000 as maximum number of generations. The grid boxes were set with 60, 60 and 60 points in the x, y and z directions, respectively. All other options were set as default. The calculated geometries were ranked in terms of free energy of binding and the best pose was selected for further analysis. Molecular visualizations were performed by Discovery Studio 4.0 client software [14].

4.8 Prediction of ADME descriptors

ADME-Tox properties of the most abundant compounds were performed by using online servers especially [http://lmmd.ecust.edu.cn:8000/predict/](http://lmmd.ecust.edu.cn:8000/predict/) and [http://preadmet.bmdrc.kr](http://preadmet.bmdrc.kr).
The List Of Abbreviations

**AD**
Alzheimer's disease

**AChE**
Acetylcholinesterase

**BuChE**
Butylcholinesterase

**CAS**
Catalytic active site

**PAS**
Peripheral anionic site

**Aβ**
Amyloid βeta

**APP**
Amyloid precursor protein

**BACE1**
β-site APP cleaving enzyme-1

**NFTs**
Neurofibrillary tangles

**IC$_{50}$**
The half maximal inhibitory concentration

**$^1$H NMR**
Proton nuclear magnetic resonance;

**$^{13}$C NMR**
Carbon-13 nuclear magnetic resonance.

Declarations

Availability of data and materials
The datasets used and analyzed during the current study are available from the corresponding author on reasonable request. We have presented all data in the form of tables and figures.

**Competing interests**

The authors declare that they have no competing interests.

**Consent for publication**

Not applicable.

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**Contributions**

M.S contributed to the design of compounds and preparation of manuscript. A.I performed docking study and contributed to preparation of manuscript. H.K.A synthesized and characterized compounds. A.R. performed the biological assay. S.N.A.B. and O.F supervised the biological tests. T.A supervised all phases of the study.

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Figures
Figure 1

Designed hybrids used to investigate as selective BuChE with Aβ aggregation inhibition, neuroprotective and metal chelating properties.
Figure 2

A: Kinetic study BuChE inhibition by compound 10h. B: Inhibition constant (Ki) of compound 10h.
Figure 3

The absorbance change of compound 10h alone and in the presence of Zn2+, Fe2+, and Cu2+ ions in the wavelength range from 200 to 600 nm.

Figure 4

Determination of the stoichiometry of complex 10h-Cu+2 using molar ratio method.
Figure 5

Schematic representation showing interactions of compound 10h with the surrounding residues of BuChE.