Accepted Manuscript

Substituted 4-morpholine *N*-arylsulfonamides as γ-secretase inhibitors

Zhiqiang Zhao, Dmitri A. Pissarnitski, Hubert B. Josien, Thomas A. Bara, John W. Clader, Hongmei Li, Mark D. McBriar, Murali Rajagopalan, Ruo Xu, Giuseppe Terracina, Lynn Hyde, Lixin Song, Lili Zhang, Eric M. Parker, Rebecca Osterman, Alexei V. Buevich

PII: S0223-5234(16)30667-5

DOI: 10.1016/j.ejmech.2016.08.024

Reference: EJMECH 8820

To appear in: *European Journal of Medicinal Chemistry*

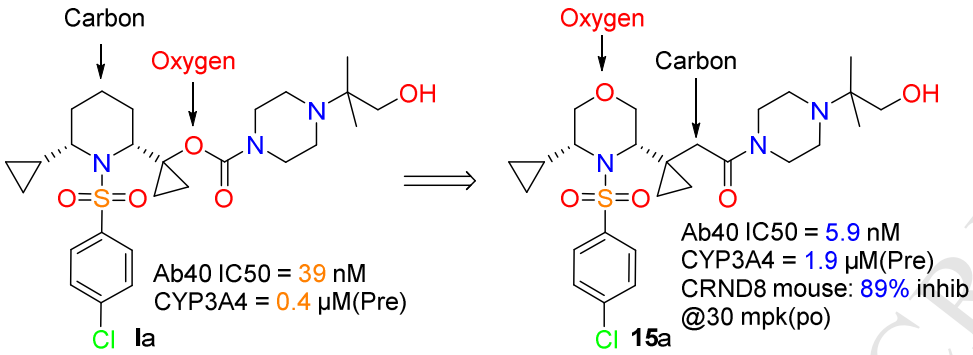
Received Date: 3 March 2016

Revised Date: 11 August 2016

Accepted Date: 12 August 2016

Please cite this article as: Z. Zhao, D.A. Pissarnitski, H.B. Josien, T.A. Bara, J.W. Clader, H. Li, M.D. McBriar, M. Rajagopalan, R. Xu, G. Terracina, L. Hyde, L. Song, L. Zhang, E.M. Parker, R. Osterman, A.V. Buevich, Substituted 4-morpholine *N*-arylsulfonamides as γ-secretase inhibitors, *European Journal of Medicinal Chemistry* (2016), doi: 10.1016/j.ejmech.2016.08.024.

Graphical Abstract



Substituted 4-morpholine N-arylsulfonamides as γ-secretase inhibitors

Zhiqiang Zhao,[a,] Dmitri A. Pissarnitski,[a] Hubert B. Josien,[a] Thomas A. Bara,[a] John W. Clader,[a] Hongmei Li,[a] Mark D. McBriar,[a] Murali Rajagopalan,[a] Ruo Xu,[a] Giuseppe Terracina,[b] Lynn Hyde,[b] Lixin Song,[b] Lili Zhang,[b] Eric M. Parker,[b] Rebecca Osterman,[c] and Alexei V. Buevich[c]*

[a] Department of Medicinal Chemistry, Merck Research Laboratories, 2000 Galloping Hill Road, Kenilworth, NJ 07033, USA

[b] Department of Neurobiology, Merck Research Laboratories, 2000 Galloping Hill Road, Kenilworth, NJ 07033, USA

[c] NMR Structure Elucidation, Merck Research Laboratories, 2000 Galloping Hill Road, Kenilworth, NJ 07033, USA

*Zhiqiang.Zhao@Merck.com

ABSTRACT: The design, synthesis, SAR, and biological profile of a substituted 4-morpholine sulfonamide series of γ-secretase inhibitors (GSIs) were described. In several cases, the resulting series of GSIs reduced CYP liabilities and improved γ-secretase inhibition activity compared to our previous research series. Selected compounds demonstrated significant reduction of amyloid-β (Aβ) after acute oral dosing in a transgenic animal model of Alzheimer's disease (AD).

Kew words: γ-secretase inhibitor; Alzheimer's disease; substituted 4-morpholine sulfonamide

1. Introduction

Alzheimer's disease (AD) is the most common form of dementia that causes problems with memory, thinking and behavior in the elderly. These problems can become so severe over time that the patients will not be able to participate in conversation and to respond to their environment. AD is the sixth leading cause of death across all ages in the United States. An estimated 5.3 million Americans have AD and causing $226 billion in cost to the nation in 2015 [1]. As the elderly population continues to grow, the prevalence of AD will increasingly be a major burden to the nation's health care system.

It is known that γ-secretase is one of the critical enzymes required for the generation of Aβ peptides. Accumulation and deposition of Aβ plaques leads to the damage and death of neurons, which is correlated with severity of AD [2]. Thus, inhibition of γ-secretase is considered to be a disease-modifying approach for this disorder [3]. Small-molecule GSIs have been shown to lower levels of plasma, cerebrospinal fluid (CSF), and cortical Aβ peptides in the animal models[4]. A review of recent advances in the identification of GSIs was published in a Journal of Medicinal Chemistry perspective [5]. The same review also discussed the application of GSIs beyond AD, such as for the treatment of leukemia [6] and breast cancer [7].

The previously published research from our group identified novel series of arylsulfonamides as γ-secretase inhibitors. Josien and Pissarnitski *et. al.* reported N-arylsulfonamides as orally active GSIs (Structure **I**, Fig. 1) [8,9]. Li *et. al.* demonstrated that adding a hydroxyl group on piperidine ring can further reduce the CYP inhibition while retain the GSI activity (Structure **II**, Fig. 1) [10].

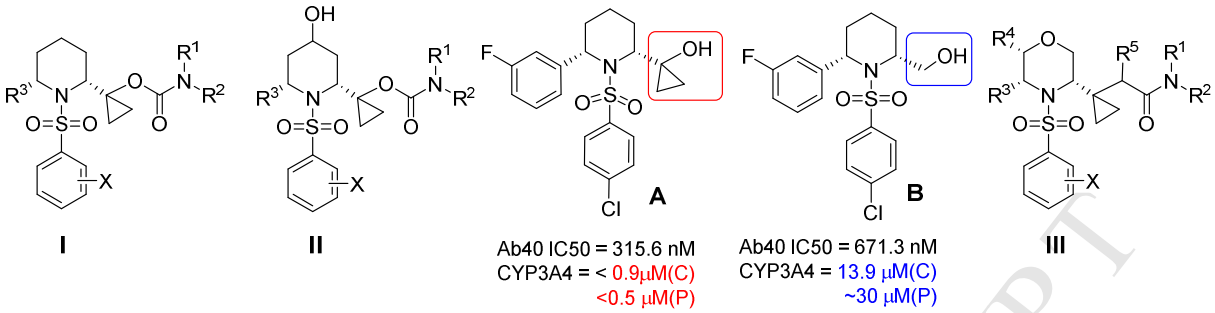


Fig. 1. Design of morpholine sulfonamide series of GSIs (**III**)

Two intermediate compounds (**A** and **B** in Fig. 1) were tested for both Aβ40 and CYP3A4 inhibition activity. Potential hydrolysis of structures **I** and **II** *in vivo* may produce **A** which is a strong CYP3A4 inhibitor. Cyclopropanol group is a suspected liability, because compound **B** without it has a clean CYP3A4 profile. However, cyclopropyl is important for γ-secretase activity. Based on these known results, herein we describe the design, synthesis, SAR, and biological profile of a novel substituted morpholine sulfonamide series of GSIs **III** [11].

Borrowing SAR elements of known sulfonamide GSIs, one carbon on the piperidine ring was replaced by oxygen; carbamate right hand side was changed to amide to avoid the potential generation of cyclopropanol group from *in vivo* hydrolysis. An optional hydroxyl group was also added at R^5 position of **III** to expand SAR by analogy to structure **II**. Selected compounds of type **III** demonstrated a significant reduction of amyloid-β (Aβ) after acute oral dosing in a transgenic animal model of AD.

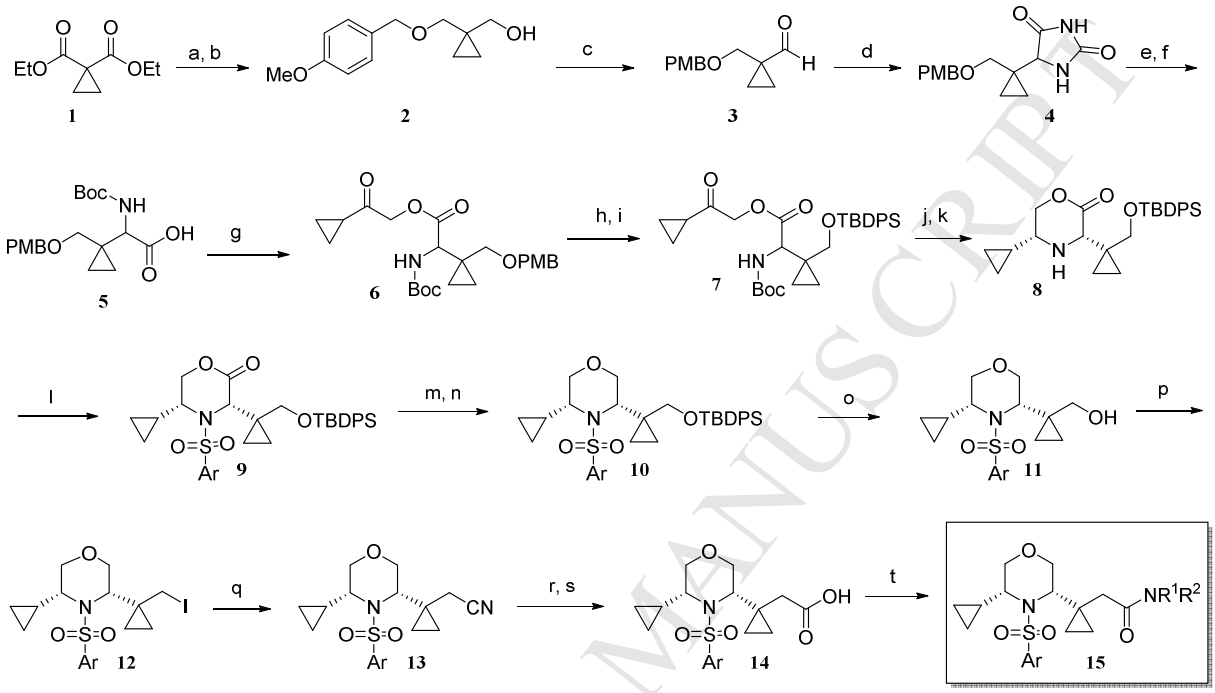
2. Results and discussion

2.1. Chemistry

2.1.1. Synthesis of compounds **15**

Our synthesis started with an inexpensive commercially available chemical **1**, diethyl cyclopropane-1,1-dicarboxylate [12] purchased from Sigma-Aldrich (Scheme 1). Compound **1** was reduced by lithium aluminum hydride (LAH) to cyclopropane-1,1-diyldimethanol [13], which was mono protected with p-methoxybenzyl (PMB) group to give (1-((4-methoxybenzyloxy)methyl)-cyclopropyl)methanol **2**. The mono-protection was moderately selective, but the bis-protected by-product could be selectively converted to the desired mono protected product **2** by treatment with one equivalent of 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ). Compound **2** was then oxidized to an aldehyde **3** by Swern oxidation. Through the Bucherer-Bergs reaction, compound **3** was converted to a hydantoin **4**. Hydrolysis of **4**, followed by protection of the resulting amine group with tert-butoxycarbonyl (Boc) gave the carboxylic acid **5**. Alkylation of **5** with 2-bromo-1-cyclopropylethanone under basic condition gave compound **6**. PMB protecting group was removed by treatment of **6** with DDQ, and the hydroxyl group was re-protected with tert-butyldiphenylsilyl (TBDPS) group to give compound **7**. Treatment of compound **7** with trifluoroacetic acid (TFA) to deprotect Boc from amine was followed by an intramolecular reaction to form a cyclic imine which was then reduced to a cyclic amine **8** by sodium triacetoxyborohydride. Sulfonylation of the amine **8** furnished sulfonamide **9**. The lactone group in compound **9** was reduced by sodium borohydride to a diol and followed by Mitsunobu reaction to give the morpholine core compound **10**. The TBDPS protecting group was removed by using tetra-n-butylammonium fluoride (TBAF) to give alcohol **11**. The hydroxyl group was then transformed to iodide **12** which was further transformed to a nitrile group **13**. Compound **13** was reduced to an aldehyde by diisobutylaluminum hydride (DIBAL-H), followed by oxidation by sodium chlorite to give the carboxylic acid **14**. The carboxylic acid **14** was then converted to a carbonyl chloride

intermediate which was used to prepare a series of final amide compounds **15a-i** by reacting with a variety of amines.



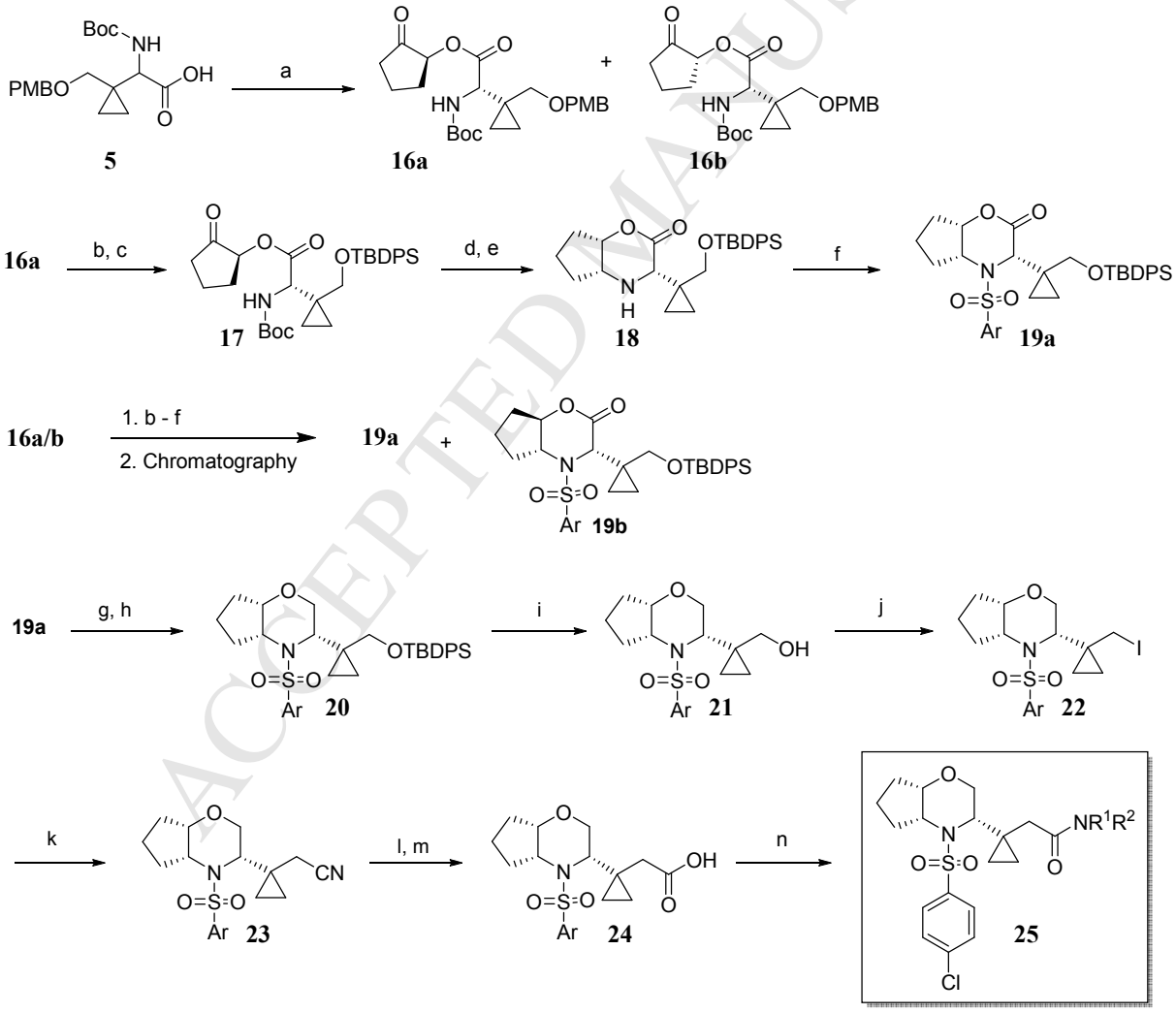
Scheme 1.[a] Synthesis of compounds **15**

[a] Reagents and conditions. (a) LAH, THF, 0°C, 87%; (b) PMBCl, NaH, DMF, 0 °C, 72%; (c) oxalyl chloride, DMSO, Et_3N, CH_2Cl_2, -78 °C – RT, 92%; (d) KCN, $(NH_4)_2CO_3$, Et_3N, water/EtOH, 60 °C, 89%; (e) $Ba(OH)_2$, water, reflux, 98%; (f) $(Boc)_2O$, Et_3N/dioxane, RT, 85%; (g) 2-bromo-1-cyclopropylethanone, KOH, KI, MeOH/CH_2Cl_2, RT, 91%; (h) DDQ, CH_2Cl_2; (i) TBDPSCl, Et_3N, imidazole, THF, 71% (two steps); (j) TFA, CH_2Cl_2; (k) $NaBH(OAc)_3$, CH_2Cl_2, 88% (two steps); (l) 4-chlorobenzene-1-sulfonyl chloride, pyridine, 51%; (m) $NaBH_4$, $CaCl_2$, THF/EtOH, 69%; (n) PPh_3, DEAD, toluene, 60%; (o) TBAF, THF, 95%; (p) I_2, Ph_3P, imidazole, CH_3CN/Toluene, 89%; (q) n-Bu_4NCN, CH_3CN, 91%; (r) DIBAL, CH_2Cl_2, -78 °C, 82%; (s) $NaClO_2$, $CH_3CHC(CH_3)_2$, t-BuOH/water, 0 °C, 97%; (t) oxalyl chloride, Et_3N, HNR^1R^2.

2.1.2. Synthesis of compound **25**

Following the same chemical route as described in Scheme 1, we used 2-chlorocyclopentanone for reaction with **5**. The intermediate compound **5** reacts with 2-chlorocyclopentanone to give a mixture of two diastereomers of compound **16** (Scheme 2). The two diastereomers of **16** were

separated by flash chromatography purification on silica gel, followed by crystallization from diethyl either. One single diastereomer **16a** (white crystal form) was carried over to compound **17**. In the cyclization step of **17** to **18**, we obtained the fused ring compound **18** which was sulfonylated to provide diastereomerically pure compound **19a** which was used to assign stereochemistry for the whole series. In order to gain additional confidence in the assignment, the alternative diastereomer **19b** was also secured by carrying a diastereomeric mixture **16a/b** through the same steps, followed by separation of two diastereomers **19a** and **19b** by flash chromatography purification on silica gel.



Scheme 2. [a] Synthesis of fused ring morpholine compounds **25**

[a] Reagents and conditions. (a) 2-Chlorocyclopentanone, KOH, KI, MeOH, RT, 70%; chromatography, crystallization; (b) DDQ, CH_2Cl_2; (c) TBDPSCl, Et_3N, imidazole, THF, 92% (two steps); (d) TFA, CH_2Cl_2, 87%; (e) $NaBH(OAc)_3$, CH_2Cl_2, 73% (two steps); (f) 4-chlorobenzene-1-sulfonyl chloride, pyridine, 71%; (g) $NaBH_4$, $CaCl_2$, THF/EtOH, 62%; (h) PPh_3, DEAD, THF, 80%; (i) TBAF, THF, 76%; (j) I_2, Ph_3P, imidazole, CH_3CN/Toluene, 90%; (k) n-Bu_4NCN, CH_3CN, 89%; (l) DIBAL, CH_2Cl_2, -78 °C, 94%; (m) $NaClO_2$, $CH_3CHC(CH_3)_2$, t-BuOH/water, 0 °C, 55%; (n) oxalyl chloride, Et_3N, HNR^1R^2

For the determination of relative stereochemistry of compounds **19a** and **19b**, the NMR signals for protons 2, 5,6,7, 11, and 13 (Fig. 2) were assigned on the basis of gCOSY, gHSQC, and NOESY (Supporting info). Compound **19a** reveals a strong NOE H5-H6, consistent with the *cis*- configuration of these protons. Furthermore, protons H7a and H7b are differentiated based on the magnitude of their NOE signal with H6 (2.4% and 1.2% respectively). Consistent with the overall shape of the molecule **19a**, H7b has a significant NOE with H13b, and H2 has a weak, but measurable NOE with H5. On the other hand, the structure of diastereomer **19b** is consistent with *trans*- configuration of protons H5 and H6, as evident by the NOE signals H5-H7b and H6-H7a respectively (Fig. 2). The assignment is re-enforced by observation of NOEs between the protons of cyclopropyl group (H11a, H12a) with proton H7b [14].

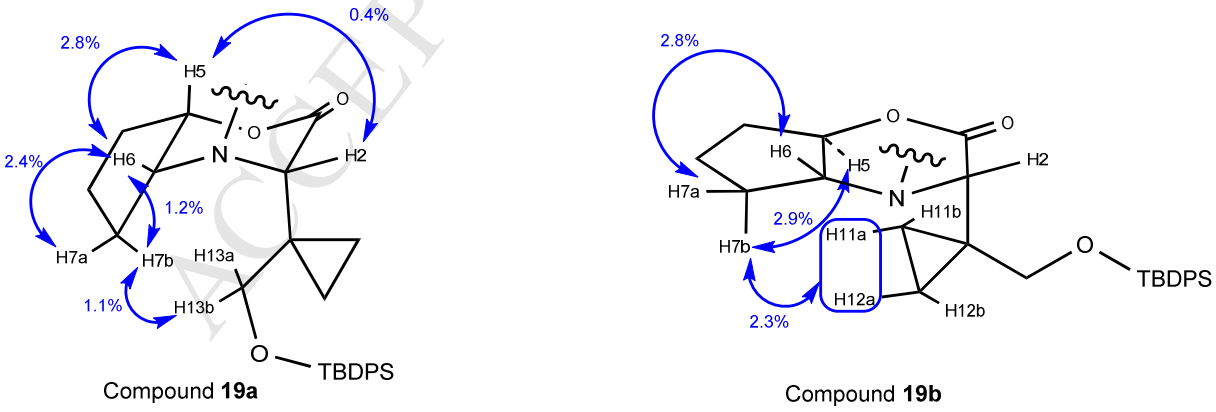
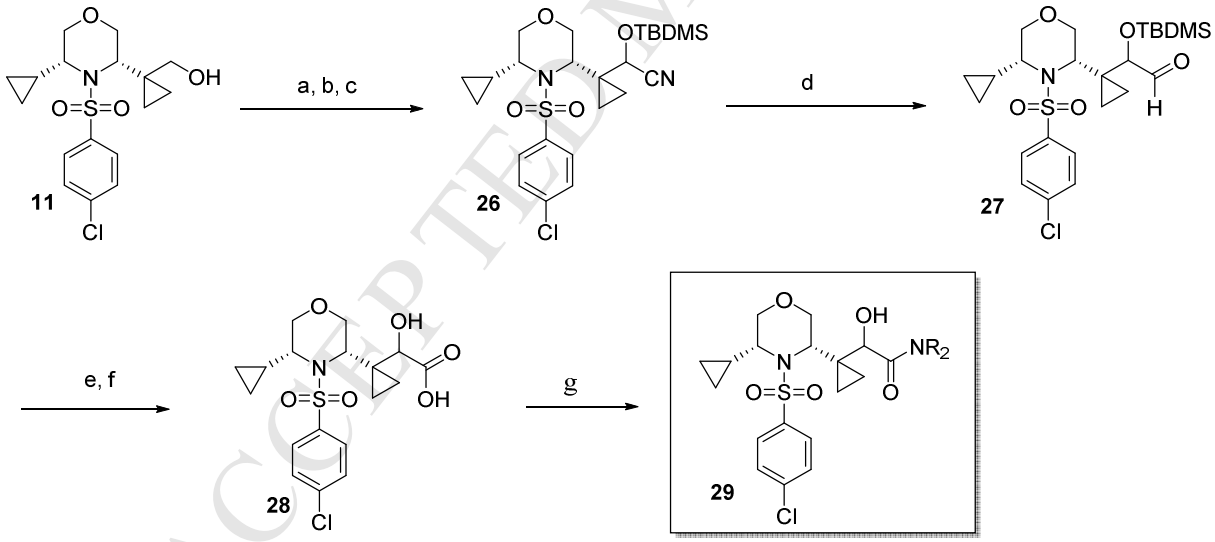


Figure 2. Assignment of structure for compounds **19a** and **19b**. 4-Clorophenylsulfonamide moiety is not shown for clarity.

Compound **19a** was transformed to the compound **21** following the same procedures in Scheme 1. Compound **21** was resolved by chiral OJ column to give the single enantiomer **21** which was carried to a series of final compounds **25a-e** (Scheme 2).

2.1.3. Synthesis of compound **29**

The intermediate compound **11** was oxidized to a corresponding aldehyde through Dess-Martin reaction (Scheme 3). Reaction of the aldehyde with KCN and TBSCl in the presence of a catalytic amount of ZnI_2 produced TBS-protected cyanohydrine **26**. The CN group in **26** was reduced by DIBAL-H to aldehyde to give compound **27**. The aldehyde group in **27** was oxidized to an acid by $NaClO_2$ and the TBS protecting group was removed by TBAF to give compound **28**. The alpha-hydroxy acid intermediate **28** was converted to the final compound **29** through the amide coupling reaction with an amine.



Scheme 3. [a] Synthesis of alpha-hydroxy amide sulfonyl morpholine

[a] Reagents and conditions. (a) DMP, CH_2Cl_2, 59%; (b) TBSCl, KCN, ZnI_2 (cat.), CH_3CN, 55 °C; (c) separation of diastereomers (d) DIBAL-H, CH_2Cl_2, -78 °C, 35%; (e) $NaClO_2$, NaH_2PO_4, $CH_3CHC(CH_3)_2$, t-BuOH/water, 87%; (f) TBAF/THF, 71%; (g) BOP, NMM, HNR^1R^2, CH_2Cl_2, 68-76%.

2.2.Biological Evaluation

2.2.1. Rational and design

Herein, we disclose the design of a series of substituted 4-morpholine sulfonamide γ-secretase inhibitors, **III** (Fig. 3), utilizing the cyclohexane framework **C** as a reference compound (Fig. 3). Although molecule **C** itself is a potent γ-secretase inhibitor *in vitro* [15], it experienced extensive

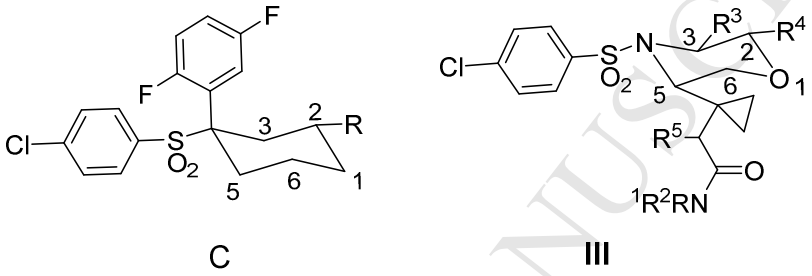


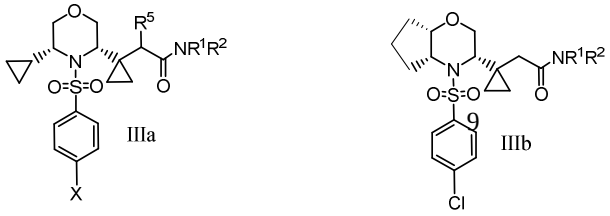
Fig. 3. Conformational comparisons for the design of GSIs.

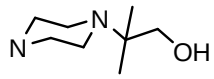
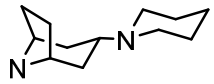
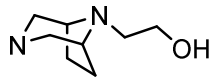
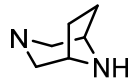
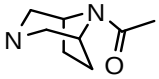
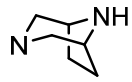
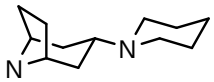
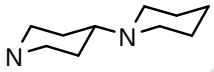
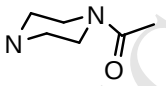
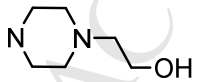
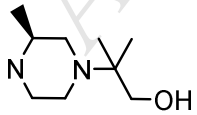
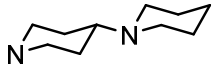
metabolism at the methylene carbons of the cyclohexyl ring in *in vitro* metabolic stability assays [16]. In addition, its poor aqueous solubility may have negatively impacted oral absorption. As discussed in our recent paper [17] the arylsulfone group must maintain the equatorial position for the potency of GSIs. Our newly substituted morpholine arylsulfonamide series maintained the active chair conformation with arylsulfonamide at equatorial position. The added substituents at C3 and C5 may help to stabilize the active chair conformation. The morpholine ring will not only increase the polarity of the molecule but may also block the *in vitro* metabolite of the methylene carbons of the cyclohexyl ring (Fig. 3).

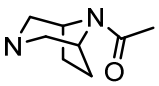
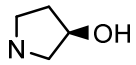
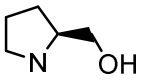
2.2.2. Biological test results

The biological test results are summarized in Table 1.

Table 1. Biological Data for compound IIIa and IIIb



Entry	*Structure NR_1R_2*	*X*	*R^5*	*Compound IIIa/IIIb*	*Cell Aβ40 IC_{50}[a] (nM)*	*CYP3A4 IC_{50} Co-inc; Pre-inc[b] (μM)*	*AUC[c] (h•ng/mL)*
1		Cl	H	**15a** (IIIa)	5.9	>30, 1.9	464
2		Cl	H	**15b** (IIIa)	12	>30, 2.0	1292
3		Cl	H	**15c** (IIIa)	6.0	>30, <0.4	2728
4		Cl	H	**15d** (IIIa)	3.5	9.9, <0.3	1160
5		Cl	H	**15e** (IIIa)	18	30, 4.3	
6		Cl	OH	**29a** (IIIa)	10	>30, 9.0	254
7		Cl	OH	**29b** (IIIa)	24	>30, >30	267
8		F	H	**15f** (IIIa)	30	>30, 1.5	575
9		F	H	**15g** (IIIa)	67	30, 17	3158
10		F	H	**15h** (IIIa)	32	>30, 6.9	1973
11		F	H	**15i** (IIIa)	14	>30, 2.2	
12		Cl	H	**25a** (IIIb)	1.8	30, 30	36
13		Cl	H	**25b** (IIIb)	1.7	21, 8.0	

14		Cl	H	**25c** (IIIb)	4.8	30, 15
15		Cl	H	**25d** (IIIb)	16	>30, >30
16		Cl	H	**25e** (IIIb)	18	30, 30

[a] Values are means of two experiments; [b] Pre-incubation values were determined 30 min after combining the compound with the enzyme; [c] Measured in rat (n = 2) over 0-6 h period after a 30 mg/kg oral dosing.

The test results show that substituted 4-morpholine sulfonamide compounds were very active γ-secretase inhibitors. The IC_{50} for the inhibition of Aβ40 are in the range of 1.7 to 67 nM (Table 1). A direct comparison of compound **15a** (entry 1, Table 1) with our previously published compound **Ia** [8] shows that **15a** is six times more potent than **Ia** in the γ-secretase assay and also has a lessened CYP3A4 liability by more than four folds (**Fig. 4)**. Compound **15a** was evaluated in a transgenic CRND8 mouse model of AD and demonstrated 89% inhibition of total Aβ in the plasma after a single oral dose of 30 mg/kg (**Fig. 5**).

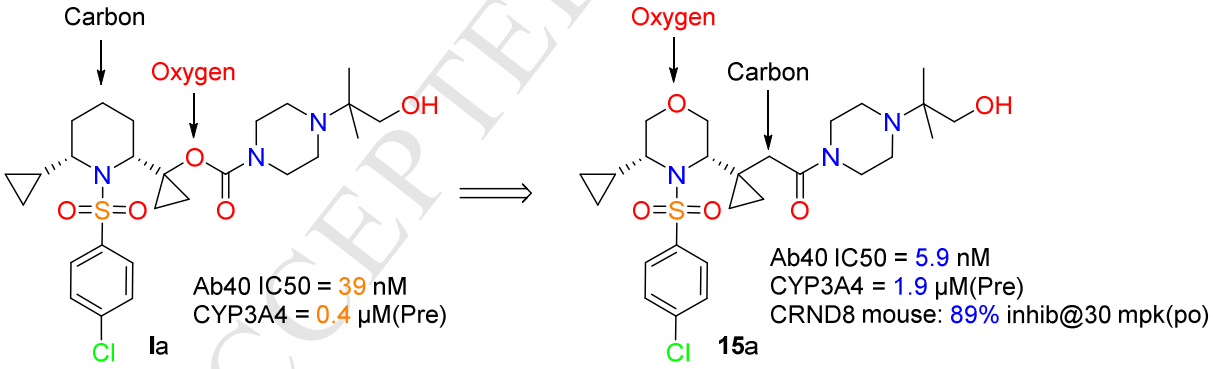


Fig. 4. Direct comparisons of **15a** and **Ia**

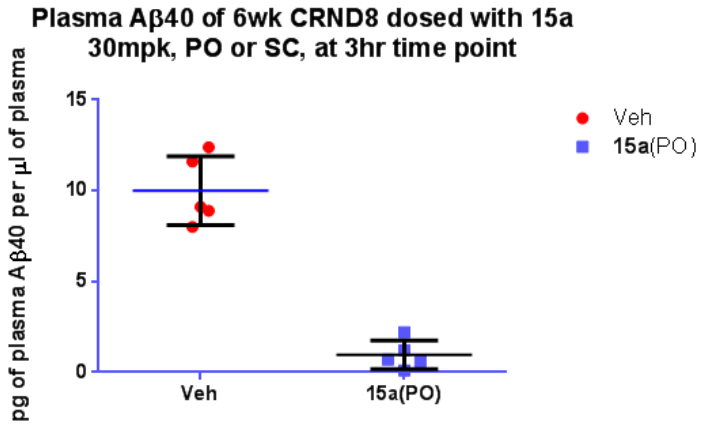


Fig. 5. Reduction of plasma Aβ40 of transgenic CRND8 mice dosed with **15a**, 30 mg/kg, PO, measured 3 h after the dosing, compared to the group treated with vehicle only (Veh).

As expected in our design, adding a hydroxy group in molecule **IIIa** at R^5 position significantly reduced CYP3A4 activity. Comparison of entries 4 and 6 shows that when R^5 changed from hydrogen to hydroxyl group, CYP3A4 activity changed from 9.9 µM to greater than 30 µM for co-incubation, and from less than 0.3 µM to 9.0 µM for pre-incubation respectively. Comparison of entry 2 and 7 demonstrates that when R^5 changed from hydrogen to hydroxyl group, CYP3A4 activity changed from 2 µM to greater than 30 µM for pre-incubation. As evident from entries 5 and 9, 4-chlorophenyl sulfonamide compound **15e** is 3 fold more potent than 4-fluorophenyl sulfonamide compound **15g**; but **15e** is also more active than **15g** for CYP3A4 inhibition. The fused ring morpholine series (**IIIb**) not only reduced the CYP3A4 activity, but also improved the γ-secretase activity. Comparison of entries 5 and 14 demonstrates that, when left side of the molecule is changed from cyclopropyl to fused cyclopentyl ring, CYP3A4 activity was ameliorated from 4.3 µM to greater than 15 µM for pre-incubation; the γ-secretase IC_{50} values improved from 18 nM to 4.8 nM. Likewise, comparison of entry 8 and 13 shows that CYP3A4 activity changed from 1.5 µM to 8.0 µM for pre-incubation, and the γ-secretase IC_{50} improved from 30 nM to 1.7 nM.

3. Conclusion

In conclusion, based on the SAR elements of known sulfonamide GSIs from our previous research results, we have designed a novel series of morpholine sulfonamide core series of GSIs. The new substituted morpholine series did not only improved the *in vitro* activity significantly, but also reduced the CYP3A4 liability. Selected compound **15a** demonstrated significant reduction of total Aβ after acute oral dosing in a transgenic animal model of AD.

4. EXPERIMENTAL

4.1. General for chemistry

Purity of all final compounds was determined by HPLC and exceeded 95% [18]. LC/MS data analyses were performed using an Applied Biosystems API-100 mass spectrometer and Shimadzu SCL-10A, LC column: Alltech platinum C18, 3 micron, 33mm x 7mm ID; gradient flow: 0 min – 10% CH_3CN, 5 min – 95% CH_3CN, 7 min – 95% CH_3CN, 7.5 min – 10% CH_3CN, 9 min – stop. The observed parent ions are given**.** Preparative chromatography was carried out on Analogix flash chromatography instrument, chiral separations were conducted on chiral OJ columns (Daicel Corp) using isopropyl alcohol and hexanes as the mobile phase. ^{1}H NMR and ^{13}C NMR spectra were obtained on either a Varian Gemini-300 (300 MHz) or XL-400 (400 MHz) and are reported as ppm down field from Me_4Si with number of protons, multiplicities, and coupling constants in Hertz indicated parenthetically. Chemical names were generated from ChemDraw. Unless noted otherwise, compounds were isolated as amorphous solids.

4.2. General for biological tests

4.2.1. *In vitro* cell based γ-secretase assay

In vitro γ-secretase assays was conducted in whole cells. HEK293 cells stably transfected with a human APP cDNA with both the Swedish and London familial AD mutations in the pcDNA3.1

vector (Invitrogen, HEK293Swe⁻Lon cells) were treated with γ-secretase inhibitors for 5 h. Aβ in conditioned medium was measured using MesoScale Discovery (MSD) technology-based sandwich immunoassays. Aβ40 was measured using the antibody pair of TAG-G2-10 and biotin-4G8. IC_{50} values for the compounds were determined as described earlier for the membrane-based assay [19]. IC_{50} values were calculated from a non-linear fit of initial reaction velocities versus compound concentration using inhibition dose-response equation (four-parameter; variable slope) using PRISM® software (GraphPad). The data presented in this study were the means of the duplicate or triplicate measurements in each experiment.

4.2.2. *In vivo* studies

All animal procedures were performed within an Association for Assessment and Accreditation of Laboratory Animal Care (AAALAC) International-accredited facility and approved by the Merck Institutional Animal Care and Use Committee (IACUC).

Transgenic 5–7 weeks old CRND8 mice carrying the Swedish and Indiana familial Alzheimer's disease APP mutations under the control of the Syrian hamster prion protein [20] used in these studies were bred at Merck Research Laboratories in Kenilworth, NJ, or Taconic in Germantown, NY, as described previously [21]. Male and female mice, counterbalanced across groups, were singly housed in a plastic igloo with nesting material before the dosing began and for the duration of the study. γ-Secretase inhibitor **15a** was formulated in 20% hydroxypropyl-β-cyclodextrin and dosed via oral gavage at a dose of 20 mg/kg. After a period of 3 hours, mice were sacrificed by CO_2 asphyxiation, total blood was collected from the posterior vena cava in EDTA Microtainer® tubes (BD Biosciences), and plasma was isolated. Plasma Aβ40 was quantified using biotin-4G8 and S-tag G2-10 using MesoScale Discovery (MSD) technology-based sandwich immunoassay.

4.2.3. Determination of rat plasma AUCs after an oral dosing

Two male Sprague-Dawley rats (Charles River, Co.) were used per each compound. The rats were pre-cannulated (femoral artery) in order to facilitate precise blood sampling times, to increase throughput, and to reduce the stress on the animals caused by serial bleedings. Rats were fasted overnight prior to oral dosing at a dose of 10 mg/kg in a 5-mL/kg dose (compounds were formulated in 0.5% methylcellulose). Blood was collected into heparin-containing tubes serially from each animal at 0.5, 1, 2, 3, 4 and 6 h post-dosing (100 μL per time point) and centrifuged to generate plasma. The plasma samples were stored at -20 °C until analysis. The concentrations of compounds in plasma was determined by a selective, but non-validated LC-MS/MS assay using selected reaction monitoring (SRM) methods developed for each compound prior to analysis of the plasma samples. The individual SRM transitions were based on a fragmentation from the protonated molecule ($[MH]^+$) to a characteristic product ion [22].

4.2.4. CYP inhibition assay

Assays for CYP inhibition were conducted using commercial recombinant 3A4, 2D6, and 2C9 enzymes (Thermo Fisher) by measuring appearance of fluorescent products from non-fluorescent substrates: 3-cyano-7-ethoxy-coumarin, dibenzyl fluorescein, 7-methoxy-4-trifluoromethylcoumarin, 7-methoxy-4-(aminomethyl)-coumarine, and 7-benzyloxy-4-trifluoromethylcoumarin. Data for the pre-incubation experiment was collected 30 min after addition of the compound to the assay system. Known CYP inhibitors, sulfaphenazole and ketoconazole were used as the positive controls. All compounds were inactive in 2D6 and 2C9 assays up to the maximal measured concentrations of 30 μM.

4.3. Procedures for synthesis of 3,5-cis disubstituted morpholine compounds **15a-i**

4.3.1. Synthesis of (1-((4-methoxybenzyloxy)methyl)cyclopropyl)methanol **2**

To solid LAH (16.6 g, 0.44 mol) in a 2 L flask at 0 °C was added dry THF (350 mL) slowly, and then starting material compound **1** (32 g, 0.202 mol) in dry THF (50 mL) was added dropwise through addition funnel at 0 ºC. Addition of the compound **1** was interrupted when too much bubbling occurred. After addition was complete, the mixture was stirred at 0 ºC for 30 minutes. The reaction was then quenched by adding water dropwise at 0 ºC with reflux condenser. The precipitate was aged for 3-4 h, filtered through celite. The precipitate was washed with 1 L of THF. The filtrate was dried ($MgSO_4$) and concentrated in vacuo to obtain a diol product (17.9 g, 87%).

To NaH (7.71 g, 193 mmol, 60% in mineral oil) in DMF (300 mL) under nitrogen at 0 ºC, was added the diol compound (17.9 g, 175 mmol) obtained from the above in DMF (200 mL) dropwise. After 30 minutes, para-methoxybenzyl chloride (PMBCl) (30.2 g, 193 mmol) in DMF (100 mL) was added dropwise over 30 minutes. The reaction was stirred at 0 ºC for 2 h. The mixture was allowed to warm up to room temperature and stirred for another 4 h. TLC analysis (30% EtOAc/Hexane) showed that the reaction was complete and had two sports. The reaction mixture was quenched with ice, extracted with diethyl ether (3×150 mL). The combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated in vacuo. The residue was purified by flash chromatography on silica gel, eluting with 0 – 30% EtOAc/Hexane in 1h to obtain the desired mono PMB protected product (22.8 g, 58.6%). ^{1}H NMR: 7.23 (2H, d, J = 8.60), 6.86 (2H, d, J = 8.62), 4.44 (2H, s), 3.79 (3H, s), 3.55 (2H, s), 3.42 (2H, s), 0.45 – 0.55 (4H, m). Mass Calcd: 222.13. m/z found: 223.3. The reaction also produce a bis PMB protected by-product (9.9 g, 16.5%). However, the bis-PMB protected by-product can be converted to the desired mono PMB protected product by the following procedure. To the by-product (9.9 g, 28.9 mmol) dissolved in a mixture of CH_2Cl_2/ water (200 mL/4 mL), was added DDQ (6.56 g, 28.9

mmol) in CH_2Cl_2 (25 mL) dropwise over a period of 30 minutes. The reaction was stirred overnight. TLC analysis (100% CH_2Cl_2) showed that the reaction was complete. The solid was filtered off and was washed with CH_2Cl_2 (3 times). The organic phases from filtrate were washed with water and brine, dried ($MgSO_4$) and concentrated in vacuo. The residue was purified by flash chromatography on silica gel, eluting with 0 – 30% EtOAc/Hexane to obtain the desired product (5.3 g, 82.5%).

4.3.2. Synthesis of compound 1-((4-methoxybenzyloxy) methyl)cyclopropanecarbaldehyde **3**

To oxalyl chloride (4.36 mL, 50 mmol) in CH_2Cl_2 (140 mL) at -78 ºC, was added DMSO (4.43 mL, 62.5 mmol) in CH_2Cl_2 (20 mL) dropwise. The mixture was stirred for 10 minutes, and then compound **2** (5.56 g, 25 mmol) in CH_2Cl_2 (70 mL) was added slowly and stirred for another 10 minutes, followed by addition of Et_3N (34.9 mL, 250 mmol, neat). Cooling bath was removed and the reaction mixture was stirred for 30 minutes. The reaction was quenched with water, extracted with CH_2Cl_2 (3 times). The combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated in vacuo. The crude product was purified by flash chromatography on silica gel, eluting with 0 - 20% EtOAc/Hexane in 1 h to afford compound **3** (5.06g, 92%). ^{1}H NMR: 9.0 (1H, s), 7.25 (2H, d, J = 8.59), 6.87 (2H, d, J = 8.43), 4.48 (2H, s), 3.80 (3H, s), 3.66 (2H, s), 1.22 (2H, m), 1.10 (2H, m). Mass Calcd: 220.11. m/z found: 221.2.

4.3.3. Synthesis of 5-(1-((4-methoxybenzyloxy)methyl)cyclopropyl)imidazolidine-2,4-dione **4**

To compound **3** (18.2 g, 0.083 mol) in 50% aqueous alcohol (100 mL) in a pressure flask, was added KCN (8.1 g, 0.124 mol) and ammonium carbonate (23.9 g, 0.25 mol) and triethylamine (50 mL, 0.35 mol). The flask was placed on the ultrasonicator for 2 minutes, and then stirred at 60 ºC overnight. TLC analysis (25%) EtOAc/Hexane showed that the reaction was complete. The reaction mixture was extracted with CH_2Cl_2 (3 times). The combined organic phases were

washed with water and brine, dried ($MgSO_4$) and concentrated. The crude product was crystallized from MeOH/CH_2Cl_2/Hexane to yield compound **4** (21.42g, 89%). ^{1}H NMR: 7.21 (2H, d, J = 8.53), 6.87 (2H, d, J = 8.66), 4.42 (2H, d, J = 2.1), 3.92 (1H, s), 3.80 (3H, s), 3.77 (1H, d, J = 9.07), 2.96 (1H, d, J = 10.3), 0.44 – 0.84 (4H, m). Mass Calcd: 290.13. m/z found: 291.3.

4.3.4. Synthesis of 2-(tert-butoxycarbonylamino)-2-(1-((4-methoxybenzyloxy)methyl)cyclopropyl) acetic acid **5**

To a solution of hydantoin compound **4** (15.56 g, 53.67 mmol) in water (200 mL) was added $Ba(OH)_2$ (18.4 g, 107.3 mmol). The flask was fitted with a reflux condenser and the mixture was refluxed overnight. TLC analysis (0.5% MeOH/ CH_2Cl_2) showed that the reaction was complete. The hot reaction mixture was filtered, then the filtrate was boiled with $(NH_4)_2CO_3$ (12.8 g) to remove barium as insoluble $BaCO_3$. Boiling was continued to decompose excess $(NH_4)_2CO_3$. The resulting filtrate was concentrated to give an amino acid intermediate (14.0g, 98%). ^{1}H NMR: 7.29 (2H, d, J = 8.75), 6.89 (2H, d, J = 8.75), 4.50 (1H, d, J = 11.37), 4.36 (1H, d, J = 11.36), 3.80 (1H, d, J = 9.83), 3.77 (3H, s), 3.15 (1H, s), 2.97 (1H, d, J = 9.83), 0.55 - 0.95 (4H, m). Mass Calcd: 265.13. m/z found:266.2.

To a mixture of amino acid intermediate (18.0 g, 0.0679 mol) in dioxane (50 mL) and water (50 mL) was added Boc_2O (26.68g, 0.122mol) and TEA (19 mL, 0.136 mol). The reaction was stirred at RT for overnight. TLC analysis (5% MeOH/ CH_2Cl_2) showed that the reaction was complete. Dioxane was removed in vacuo. The mixture was extracted with EtOAc (3 times), and organic phase was washed with citric acid (20%). The aqueous phase was back extracted with EtOAc. The combined organic phases were dried ($MgSO_4$) and concentrated. The crude product was purified by flash chromatography on silica gel, eluting with 0-15% MeOH/CH_2Cl_2 to yield

the compound **5** (21.0g, 85%). ^{1}H NMR: 7.25 (2H, d, J = 8.68), 6.87 (2H, d, J = 8.64), 6.06 (1H, bs), 4.55 (1H, d, J = 11.16), 4.43 (1H, d, J = 11.32), 3.95 (1H, m), 3.80 (3H, s), 2.89 (1H, d, J = 9.94), 0.805 (1H, m), 0.64 (2H, t, J = 6.64), 0.55 (1H, m). Mass Calcd: 365.18. m/z found: 366.4.

4.3.5. Synthesis of 2-cyclopropyl-2-oxoethyl 2-(tert-butoxycarbonylamino)-2-(1-((4-methoxybenzyloxy) methyl)cyclopropyl)acetate **6**

To a solution of compound **5** (21.38g, 0.0737mol) in MeOH (150 mL) and CH_2Cl_2 (50 mL) was added slowly a solution of KOH (4.14g, 0.0737 mol) in MeOH (150 mL). The reaction is concentrated after 2 minutes. The mixture was concentrated for several times with CH_2Cl_2 to remove any trace amount of MeOH as much as possible. The resulting solid and KI (1.22g, 7.37mmol) are taken up in DMF (50 mL) and a solution of 2-bromo-1-cyclopropylethanone (12.0 g, 0.0737 mol) in DMF (25 mL) was slowly added (dark brown color to yellow with precipitate). The reaction mixture was stirred at RT for 4 h. TLC analysis (30% EtOAc/Hexane) showed that the reaction was complete. The reaction mixture was extracted with ether (3 times). The combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated in vacuo. The crude product was purified by flash chromatography on silica gel, eluting with 0 to 30% EtOAc/Hexane to give compound **6** (29.9g, 91%). ^{1}H NMR: 7.27 (2H, d, J = 8.66), 6.86 (2H, d, J = 8.66), 6.37 (1H, d, J = 7.92), 4.95 (1H, d, J = 16.6), 4.72 (1H, d, J = 16.6), 4.49 (1H, d, J = 11.2), 4.35 (1H, d, J = 11.2), 3.95 (1H, d, J = 11.4), 3.80 (3H, s), 3.67 (1H, d, J = 7.84), 2.88 (1H, d, J = 10.34), 1.98 (1H, m), 1.58 (1H, s), 1.43 (9H, s), 1.09 (1H, m), 0.96 (2H, m), 0.89 (1H, m), 0.79 (1H, m), 0.64 (1H, m), 0.57 (1H, m). Mass Calcd: 447.23. m/z found: 448.3.

4.3.6. Synthesis of 2-cyclopropyl-2-oxoethyl 2-(tert-butoxycarbonylamino)-2-(1-((tert-butyldiphenylsilyloxy) methyl) cyclopropyl)acetate **7**

To compound **6** (30.2 g, 0.0675 mol) in CH_2Cl_2 (400 mL) and water (8 mL), was added DDQ (16.85 g, 0.0742 mol) as solid in portions over a period of 30 minutes. The reaction was stirred overnight. TLC analysis (100% CH_2Cl_2, use stain) showed that the reaction was complete. The solid was filtered off and washed with CH_2Cl_2 (3 times). The combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated. The residue was purified by flash chromatography on silica gel to collect all fractions, eluting with 0 to 35% EtOAc/Hexane to obtain product (22.1 g, 100%, not pure, taken as is).

To a solution of the above product (22.1 g, 67.5 mmol) in THF (160 mL), was added imidazole (9.19 g, 135 mmol), followed by TBDPSCl (21 mL, 8 1mmol). The reaction was stirred overnight. TLC analysis (25% EtOAc/Hexane) showed that the reaction was complete (less polar, UV active product). The mixture was extracted with ether (3 times). The combined organic phases were washed with water, saturated $NaHCO_3$ and brine, dried $MgSO_4$ and concentrated. The crude product was purified by flash chromatography on silica gel, eluting with (30%-35% EtOAc/Hexane) to afford compound **7** (27.0g, 71%). ^{1}H NMR: 7.67 (4H, m), 7.39 (6H, m), 6.81 (1H, d, J = 7.78), 4.98 (1H, d, J = 16.57), 4.70 (1H, d, J = 16.56), 4.28 (1H, d, J = 11.18), 3.69 (1H, d, J = 7.72), 2.83 (1H, d, J = 10.59), 1.99 (1H, m), 1.44 (9H, s), 1.08 (9H, s), 0.94 (3H, m), 0.81 (3H, m), 0.53 (1H, m), 0.41 (1H, m). Mass Calcd: 565.29. m/z found: 566.5.

4.3.7. Synthesis of (3S,5R)-3-(1-((tert-butyldiphenylsilyloxy)methyl)cyclopropyl)-5-cyclopropylmorpholin-2-one **8**

To a solution of **7** (27.0 g, 47.76 mmol) in CH_2Cl_2 (270 mL) was added TFA (90 mL) and stirred for 2 h. TLC analysis (10% EtOAc/Hexane) showed that the reaction was complete. Solvent was removed. The residue was redissolved in CH_2Cl_2 and toluene, solvent was removed in vacuo. This step was repeated two more times to remove TFA. The residue was dissolved in

CH_2Cl_2 (250 mL), washed with aqueous $NaHCO_3$, water and brine, dried ($MgSO_4$) and concentrated to yield the cyclized imine product (22.1 g, 100%, crude).

To a solution of the cyclized imine product (12.6 g, 28.17 mmol) in CH_2Cl_2 (280 mL) was added $NaBH(OAc)_3$ (11.94 g, 56.34 mmol). The reaction was stirred at RT overnight. The reaction mixture was diluted with saturated $NaHCO_3$, extracted with CH_2Cl_2 (3 times). The combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated. The crude product was purified by flash chromatography on silica gel, eluting with 0 to 35% EtOAc/Hexane to afford compound **8** (11.2 g, 88%). ^{1}H NMR: 7.70 (4H, m), 7.40 (6H, m), 4.31 (1H, dd, J = 3.38, 10.52), 4.21 (1H, t, J = 10.30), 3.88 (1H, d, J = 10.55), 3.48 (1H, d, J = 10.56), 3.07 (1H, s), 2.27 (1H, m), 1.05 (9H, s), 0.87 (1H, m), 0.27 – 0.69 (8H, m). ^{13}C NMR (Chloroform-*d*) δ (ppm): 169.30, 135.69, 135.64, 133.44, 133.28, 129.67, 129.64, 127.65, 127.64, 77.25, 76.75, 73.42, 67.52, 63.57, 57.25, 26.78, 24.85, 19.19, 12.25, 10.54, 7.99, 2.75, 1.99. Mass Calcd: 449.24. m/z found: 450.4.

4.3.8. Synthesis of (3S,5R)-3-(1-((tert-butyldiphenylsilyloxy)methyl)cyclopropyl)-4-(4-chlorophenylsulfonyl)-5-cyclopropylmorpholin-2-one **9**

To a solution of starting compound **8** (4.20 g, 9.35 mmol) in pyridine (80.0 mL) was added 4-chlorobenzene-1-sulfonyl chloride as solid. The reaction was stirred at 85 ºC overnight. The reaction was diluted with aqueous $NaHCO_3$, extracted with CH_2Cl_2 (3 times), the combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated. The residue was purified by flash chromatography on silica gel, eluting with 0 to 30% EtOAc/Hexane in 1 h to afford compound **9** (7.39g, 51.0%). ^{1}H NMR: 7.63 (6H, m), 7.41 (8H, m), 4.68 (1H, s), 4.54 (1H, dd, J = 10.11, 12.48), 4.11 (1H, dd, J = 5.75, 12.07), 3.74 (1H, d, J = 10.99), 3.55 (1H, d, J = 10.95), 2.91 (1H, m), 1.03 (9H, s), 0.90 (1H, ,m), 0.82 (1H, m), 0.5 – 0.65 (6H, m), 0.21 (1H, m). Mass Calcd: 623.19. m/z found: 624.1.

4.3.9. Synthesis of (3S,5R)-3-(1-((tert-butyldiphenylsilyloxy)methyl)cyclopropyl)-4-(4-chlorophenylsulfonyl)-5-cyclopropylmorpholine **10**

To starting compound **9** (10.35 g, 16.58 mmol) and $CaCl_2$ (11.0 g, 99.0 mmol) in THF (85.0 mL) and EtOH (127 mL) was added $NaBH_4$ (3.14 g, 83.0 mmol) as solid. The reaction was stirred at RT for 2 h. TLC analysis (25% EtOAc/Hexane) showed that the reaction was incomplete. Stirring was continued until the less polar product was converted completely to the more polar product. At 0 ºC, the reaction was quenched (exothermic) with citric acid (20%), extracted with CH_2Cl_2 (3 times). The combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated. The crude product was purified by flash chromatography on silica gel, eluting with 0 to 30% EtOAc/Hexane in 1 h to afford the desired diol product (7.14g, 69.1%). ^{1}H NMR: 7.77 (2H, d, J = 8.84), 7.62 – 7.55 (4H, m), 7.46 – 7.36 (8H, m), 4.10 (1H, d, J = 10.26, 3.86 (1H, t, J = 10.11), 3.80 (2H, d, J = 6.01), 3.60 (1H, s), 3.40 (1H, t, 7.08), 2.86 (1H, d, J = 11.70), 2.75 (1H, q, J = 9.87), 0.95 (1H, m), 0.87 (1H, m), 0.67 (3H, m), 0.13 – 0.29 (3H, m), 0.04 (1H, m). Mass Calcd:627.22. m/z found:628.3.

To the diol starting material (7.14 g, 11.38 mmol) obtained from above and PPh_3 (6.57 g, 25.04 mmol) in toluene (75.0 mL) was added DEAD (3.96 mL, 25.04 mmol) at RT. The reaction was stirred at RT overnight. TLC analysis (25% EtOAc/Hexane) showed that the reaction was complete with formation of a less polar product. The reaction mixture was diluted with water, extracted with CH_2Cl_2 (3 times). The combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated. The crude product was purified by flash chromatography on silica gel, eluting with 0 to 35% EtOAc/Hexane to afford the product **10** (4.2 g, 59.7%). ^{1}H NMR: 7.63 (2H, d, J = 8.81), 7.61 – 7.66 (4H, m), 7.40 (6H, m), 7.32 (2H, d, J = 8.87), 4.19 (1H, d, J = 4.29), 4.01 (1H, d, J = 10.16), 3.88 (1H, d, J = 10.16), 3.70 (1H, d, J =

11.38), 3.55 (1H, d, J = 12.41), 3.11 (2H, m), 2.61 (1H, m), 1.05 (9H, s), 0.83 (3H, m), 0.45 (3H, m), 0.11 (3H, m). Mass Calcd: 609.21. m/z found: 610.3.

4.3.10. Synthesis of (1-((3S,5R)-4-(4-chlorophenylsulfonyl)-5-cyclopropylmorpholin-3-yl)cyclopropyl)methanol **11**

To a starting compound **10** (4.2 g, 6.88 mmol) in THF (50 mL) was added TBAF (1 M in THF, 13.76 mmol) at RT. The reaction was stirred at RT for 2 h. TLC analysis (80% CH_2Cl_2/Hexane) showed that the reaction was complete. The reaction mixture was quenched with water, extracted with EtOAc (3 times). The combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated. The crude product was purified by flash chromatography on silica gel, eluting with 0 to 30% EtOAc/Hexane to obtain desired product **11** (2.43 g, 95%). Compound **11** was further resolved by HPLC Chiral OJ column to obtain a single enantiomer. $[\alpha]_D^{20}$ = -125.84 (EtOH). ^{1}H NMR: 7.75 (2H, d, J = 8.52), 7.52 (2H, d, J = 8.52), 4.68 (1H, dq, J = 12.44, 2.2), 4.17 (1H, d, J = 4.28), 3.74 (1H, d, J = 11.50), 3.67 (1H, dd, J = 4.59, 10.43), 3.32 (1H, d, J = 12.61), 3.01 (1H, dd, J = 3.75, 11.46), 2.85 (1H, dd, J = 3.75, 10.54), 2.78 (1H, dd, J = 4.44, 12.64), 2.64 (1H, dd, J = 10.49, 12.34), 1.65 (1H, m), 1.25 (1H, m), 0.51 – 0.81 (6H, m), 0.28 (1H, m). Mass Calcd: 371.10. m/z found: 372.3.

4.3.11. Synthesis of (3R,5S)-4-(4-chlorophenylsulfonyl)-3-cyclopropyl-5-(1-(iodomethyl)cyclopropyl)morpholine **12**

The starting hydroxyl compound **11** (0.478 g, 1.29 mmol) in CH_3CN (4 mL) and toluene (8 mL) was treated with Ph_3P (406 mg, 1.55 mmol), I_2 (393 mg, 1.55 mmol) and imidazole (263.5 mg, 3.87 mmol) at RT. The reaction was stirred at RT for 1 h. TLC analysis (30% EtOAc/Hexane) showed that the reaction was complete. The reaction mixture was quenched with NH_4Cl, extracted with EtOEt (3 times). The combined organic phases were washed with saturated $NaHCO_3$ and brine, dried ($MgSO_4$) and concentrated. The residue was purified by flash

chromatography on silica gel, eluting with 0 to 80% CH_2Cl_2/Hexane in 1h to afford the desired product (0.55g, 89%). ^{1}H NMR: 7.82 (2H, d, J = 8.83), 7.51 (2H, d, J = 8.83), 4.33 (1H, d, J = 3.95), 4.12 (1H, d, J = 10.25), 3.74 (1H, d, J = 11.45), 3.46 (1H, d, J = 12.79), 3.17 (1H, dd, J = 3.88, 11.45), 3.13 (1H, d, J = 10.10), 3.09 (1H, dd, 4.30, 12.73), 2.73 (1H, dd, J = 3.67, 10.27), 1.97 (1H, m), 1.61 (1H, m), 0.99 (1H, m), 0.84 (1H, m), 0.72 (1H, m), 0.62 (1H, m), 0.52 (1H, m), 0.38 (1H, m), 0.22 (1H, m). Mass Calcd: 481.00. m/z found: 482.1.

4.3.12. Synthesis of 2-(1-((3S,5R)-4-(4-chlorophenylsulfonyl)-5-cyclopropylmorpholin-3-yl)cyclopropyl)acetonitrile **13**

To a suspension of iodide **12** (0.55 g, 1.14 mmol) in CH_3CN (10 mL, not soluble) was added n-Bu_4NCN (371 mg, 1.38 mmol) at RT. The reaction mixture was stirred at RT for 1.5 h. TLC analysis (70% CH_2Cl_2/Hexane) showed that the reaction was complete. The reaction mixture was diluted with water, and extracted with EtOAc (3 times). The combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated. The crude product was purified by flash chromatography, eluting with 0 to 30% EtOAc/Hexane in 40 minutes to give the compound **13** (0.393g, 91%). ^{1}H NMR: 7.71 (1H, d, J = 8.77), 7.51 (1H, d, J = 8.77), 4.06 (1H, d, J = 4.17), 3.72 (1H, d, J = 11.47), 3.34 (1H, d, J = 12.81), 3.14 (2H, q, J = 17.46), 2.95 (1H, dd, J = 3.74, 11.49), 2.86 (1H, dd, J = 4.35, 12.81), 2.78 (1H, dd, J = 3.43, 10.39), 1.63 (1H, m), 1.36 (1H, m), 0.91 (2H, m), 0.62 – 0.75 (4H, m), 0.30 (1H, m). Mass Calcd: 380.1. m/z found: 381.4.

4.3.13. Synthesis of 2-(1-((3S,5R)-4-(4-chlorophenylsulfonyl)-5-cyclopropylmorpholin-3-yl)cyclopropyl)acetic acid **14**

To the nitrile starting material **13** (0.393 g, 1.03 mmol) in CH_2Cl_2 (7 mL) was added DIBAL-H (1.55 mL, 1.55 mmol) at -78 ºC dropwise. The reaction was stirred at -78 ºC for 4 h. TLC analysis (10% EtOAc/Hexane) showed that the reaction was complete. The reaction mixture was

quenched by slow addition of MeOH (2 mL) and stirred for 10 minutes. H_2SO_4 (1N, 2 mL) was added and stirring continued for 45 minutes. Reaction mixture was extracted with CH_2Cl_2 (3 times), the combined organic phases were washed with water, brine, dried ($MgSO_4$) and concentrated. The crude product was purified by flash chromatography on silica gel, eluting with 0 to 20% EtOAc/Hexane to afford the aldehyde intermediate product (0.325g, 82%). ^{1}H NMR: 9.83 (1H, s), 7.69 (2H, d, J = 8.81), 7.48 (2H, d, J = 8.81), 4.12 (1H, d, J = 4.40), 3.75 (1H, dq, J = 18.43, 0.80), 3.73 (1H, d, J = 11.30), 3.36 (1H, d, J = 12.72), 3.01 (1H, dd, J = 3.78, 11.45), 2.84 (1H, dd, J = 3.60, 10.39), 2.78 (1H, dd, J = 4.43, 12.72), 1.83 (1H, d, J = 17.77), 1.72 (1H, m), 1.29 (1H, m), 0.72 – 0.83 (2H, m), 0.65 (3H, m), 0.47 (1H, m), 0.28 (1H, m). Mass Calcd: 383.10. m/z found: 384.2.

To the aldehyde intermediate (0.325g, 0.85mmol) obtained above in t-BuOH (12 mL) and water (3 mL) at 0 ºC, was added 2-methyl-2-butene (361ul, 3.4 mmol), followed by sodium chlorite (246 mg, 2.72 mmol). The reaction was stirred at RT for 2 h. TLC analysis (30% EtOAc/Hexane) showed that the reaction was complete. The reaction was quenched by saturated NH_4Cl (10 mL) and extracted with EtOAc (3×25 mL). The combined organic phases were washed with water, brine, dried ($MgSO_4$) and concentrated. The crude product was purified by flash chromatography on silica gel, eluting with 0 to 30% EtOAc/Hexane to afford **14** (330 mg, 97%). ^{1}H NMR: 7.72 (2H, d, J = 8.63), 7.46 (2H, d, J = 8.39), 4.24 (1H, d, J = 4.29), 3.73 (1H, d, J = 11.74), 3.56 (1H, dd, J = 1.52, 17.72), 3.36 (1H, d, J = 12.74), 3.04 (1H, dd, J = 3.67, 11.32), 2.82 (2H, td, J = 12.55, 4.34), 1.92 (1H, d, J = 17.75), 1.72 (1H, m), 1.25 (1H, m), 0.63 – 0.79 (5H, m), 0.55 (1H, m), 0.27 (1H, m). Mass Calcd: 399.09. m/z found: 400.2.

4.3.14. General procedure for synthesis of final compounds **15a-i**

To the carboxylic acid **14** (60 mg, 0.15 mmol) in CH_2Cl_2 (2 mL) was added oxalyl chloride (105 µl, 1.2 mmol), stirred for 20 min. Solvent was removed and the residue was placed on the high vacuum for 1h. The crude product was dissolved in 1 mL of CH_2Cl_2 and added to a vial contains amine (0.30 mmol) and Et_3N (126 µl, 0.9 mmol) in CH_2Cl_2 (1 mL). The reaction was stirred overnight. The reaction was diluted with aqueous $NaHCO_3$, extracted with CH_2Cl_2 (3 times). The combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated. The crude was purified by preparative TLC (7 % MeOH/ CH_2Cl_2) to obtain the final product.

Compound **15a**. Yield: 96%. ^{1}H NMR: 7.70 (2H, d, J = 8.74), 7.45 (2H, d, J = 8.76), 4.31 (1H, s), 3.68 (1H, d, J = 11.38), 3.35 – 3.65 (6H, m), 2.91 (1H, dd, J = 3.80, 11.41), 3.34 (2H, s), 2.74 (4H, m), 2.63 (1H, m), 2.50 (2H, m), 1.77 (1H, m), 1.70 (1H, d, J = 16.45), 1.27 (1H, m), 1.02 (6H, s), 0.63 – 0.83 (5H, m), 0.48 (1H, m), 0.28 (1H, m). ^{13}C NMR (Chloroform-*d*) δ (ppm): 169.07, 139.60, 139.31, 129.88, 128.09, 68.51, 67.71, 66.12, 63.67, 58.46, 52.97, 46.12, 42.69, 38.85, 38.59, 20.32, 18.14, 15.04, 12.56, 11.16, 6.36, 4.80. Mass Calcd: 539.22. m/z found: 540.3.

Compound **15b**. Yield: 71 %. ^{1}H NMR: 7.71 (2H, m), 7.45 (2H, m), 4.70 (0.5 H, t, J = 6.60), 4.63 (0.5 H, t, J = 5.88), 4.46 (0.5H, d, J = 4.31), 4.29 (0.5 H, d, J = 4.23), 4.18 (0.5 H, t, J = 6.98), 4.05 (0.5 H, t, J = 5.37), 3.67 (1H, dd, J = 3.38, 11.42), 3.43 (2H, m), 2.80 (4H, m), 2.48 (4H, m), 1.51 – 2.15 (13H, m), 1.39 (2H, m), 1.25 (1H, m), 0.71 (6H, m), 0.52 (1H, m), 0.26 (1H, m). Mass Calcd: 575.26. m/z found: 576.1.

Compound **15c**. Yield: 40%. ^{1}H NMR: 7.72 (1H, d, J = 8.66), 7.69 (1H, d, J = 8.67), 7.45 (1H, d, J = 8.71), 7.44 (1H, d, J = 8.67), 4.44 (0.5H, d, J = 4.21), 4.23 (1H, m), 4.08 (0.5 H, d, J = 12.92), 3.66 (2H, m), 3.58 (2H m), 3.49 (1H, m), 3.38 (1H, m), 3.23 (3H, m), 2.72 – 2.96 (4H,

m), 2.53 (3H, m), 1.46 – 1.96 (5H, m), 1.27 (1H, m), 0.62 – 0.85 (6H, m), 0.45 (1H, m,), 0.28 (1H, m). Mass Calcd: 537.21. m/z found: 538.1.

Compound **15d**. Yield: 64 %. ^{1}H NMR: 7.71 (2H, m), 7.46 (2H, m), 4.44 (0.5H, d, J = 4.30), 4.14 – 4.29 (1.5 H, m), 3.35 – 3.70 (5H, m), 2.74 – 2.94 (3H, m), 1.61 – 2.06 (9H, m), 1.28 (1H, m), 0.62 – 0.86 (6H, m), 0.47 (1H, m), 0.28 (1H, m). Mass Calcd: 493.18. m/z found: 494.3.

Compound **15e**. Yield: 55 %. ^{1}H NMR: 7.68 (2H, m), 7.46 (2H, m), 4.75 (1H, d, 15.45), 4.45 (1H, t, J = 4.6), 4.17 (2H, m), 3.68 (2H, m), 3.2 – 3.59 (3H, m), 2.72 – 2.96 (5H, m), 2.11 (3H, s), 2.13 (2H, d, J = 1.4), 2.04 (1H, d, J = 6.60), 1.76 (1H, m), 1.30 (1H, m), 0.60 – 0.90 (6H, m), 0.45 (1H, m), 0.29 (1H, m). Mass Calcd: 535.19. m/z found: 536.3.

Compound **15 f**. Yield: 72%. ^{1}H NMR: 7.78 (2H, m), 7.16 (2H, m), 4.74 (0.5 H, d, J = 13.67), 4.67 (0.5 H, d, J = 13.63), 4.37 (0.5 H, d, J = 3.98), 4.27 (0.5 H, d, J = 4.22), 3.89 (1H, t, J = 14.31), 3.56 – 3.71 (2H, m), 3.37 (1H, d, J = 12.53), 3.07 (0.5H, t, J = 12.80), 2.91 (1.5 H, m), 2.77 (2H, m), 2.49 (5H, m), 1.78 (6H, m), 1.56 (4H, m), 1.41 (2H, m), 1.26 (1H, m), 0.64 – 0.81 (6H, m), 0.47 (1H, m), 0.27 (1H, m). Mass Calcd: 533.27. m/z found: 534.1.

Compound **15g**. Yield: 74%. ^{1}H NMR: 7.76 (2H, m), 7.16 (2H, m), 4.30 (1H, m), 3.92 (1H, m), 3.34 – 3.80 (11H, m), 2.90 (1H, m), 2.76 (2H, m), 2.09 (3H, s), 1.78 (2H, s), 1.60 (1H, dd, J = 6.90, 16.73), 1.29 (1H, m), 0.85 (1H, m), 0.75 (2H, m), 0.65 (2H, d, J = 7.85). Mass Calcd: 493.20. m/z found: 494.1.

Compound **15h**. Yield: 87%. ^{1}H NMR: 7.78 (2H, dd, J = 5.05, 8.82), 7.16 (2H, t, J = 8.50), 4.31 (1H, d, J = 4.19), 3.54 – 3.76 (7H, m), 3.44 (1H, m), 3.38 (1H, d, J = 12.69), 2.90 (1H, dd, J = 3.81, 11.44), 2.75 – 2.79 (2H, m), 2.67 (1H, m), 2.57 (4H, t, J = 5.25), 2.47 (2H, m), 1.77 (1H, m), 1.71 (1H, d, J = 16.45), 1.28 (1H, m), 0.64 – 0.83 (5H, m), 0.47 (1H, m). Mass Calcd: 495.22. m/z found: 496.3.

Compound **15i**. Yield: 63%. ^{1}H NMR: 7.77 (2H, m), 7.15 (2H, t, J = 8.48), 4.00 – 4.84 (2H, m), 3.30 – 3.71 (6H, m), 2.60 – 2.97 (6H, m), 2.15 – 2.51 (2H, m), 1.74 (3H, m), 1.22 – 1.48 (4H, m), 1.03 (3H, s), 0.97 (3H, s), 0.77 (2H, m), 0.64 (3H, m), 0.47 (1H, m), 0.27 (1H, m). Mass Calcd: 537.27. m/z found: 538.3.

4.4. Procedures for synthesis of fused ring morpholine compounds **25a-e**

4.4.1. Synthesis of 2-oxocyclopentyl 2-(tert-butoxycarbonylamino)-2-(1-((4-methoxybenzyloxy) methyl)cyclopropyl) acetate **16**

The same procedure as in the synthesis of compound **6** was used for compound **16**. The product was purified by flash chromatography, eluting with 0 – 30% EtOAc/Hexane to give a mixture of two diastereomers (Total yield 71.0%). The mixture was separated by crystallization from hot diethyl ether to give a white crystal and a yellow oil compound with a 1:1 ratio of two diastereomers. Diastereomer one **16a**.(35%, white solid, mp. 63-66 °C). ^{1}H NMR: 7.27 (2H, d, J = 8.66), 6.86 (2H, d, J = 8.66), 6.25 (1H, d, J = 7.69), 5.02 (1H, t, J = 8.80), 4.79 (1H, s), 4.47 (1H, d, J = 11.27), 4.35 (1H, d, J = 11.25), 3.79 (3H, s), 3.64 (1H, d, J = 7.84), 2.93 (1H, d, J = 10.38), 2.29 (2H, m), 2.08 (1H, m), 1.84 (2H, m), 1.43 (9H, s), 0.87 (1H, m), 0.78 (1H, m), 0.60 (1H, m), 0.54 (1H, m). Mass Calcd: 477.23. m/z found: 478.3. Diastereomer two **16b** (36%, yellow oil). ^{1}H NMR: 7.29 (2H, d, J = 8.56), 6.87 (2H, d, J = 8.36), 6.35 (1H, d, J = 8.06), 5.21 (1H, t, J = 10.03), 4.53 (1H, d, J = 11.22), 4.35 (1H, d, J = 11.24), 4.01 (1H, d, J = 10.28), 3.80 (3H, s), 3.64 (1H, d, J = 8.13), 3.47 (1H, q, J = 7.55, 14.03), 2.84 (1H, d, J = 10.39), 2.21 – 2.41 (3H, m), 2.09 (1H, m), 1.87 (2H, m), 1.44 (9H, s), 1.21 (1H, t, J = 7.02), 0.79 (2H, m), 0.63 (1H, m), 0.53 (1H, m). Mass Calcd: 477.23. m/z found: 478.3.

4.4.2 Synthesis of 2-oxocyclopentyl 2-(tert-butoxycarbonylamino)-2-(1-((tert-butyldiphenylsilyloxy) methyl)cyclopropyl) acetate **17**

To compound **16a** (18.0 g, 40.2 mmol) in DCM (214 mL) and water (4.5 mL) was added DDQ (10 g, 44.2 mmol) as solid slowly by portion. Then the reaction was stirred at RT overnight. TLC analysis (100% CH_2Cl_2, use stain) showed that the reaction was done (polar, non UV active product). The solid was filtered off and filtrate was partitioned between water and CH_2Cl_2. The organic phase was washed with water and brine, dried ($MgSO_4$) and concentrated. The residue was purified by flash chromatography on silica gel, eluting with 0 - 35% EtOAc/Hexane to afford the product (13.2 g, 100%).

To the resulting hydroxyl product (13.2 g, 40.2 mmol) in THF (100 mL) was added imidazole (5.49 g, 68.1 mmol), followed by TBDPSCl (13.3 g, 48.4 mmol). The reaction was stirred at RT overnight. TLC (25% EtOAc/Hexane) analysis showed that the reaction was complete. The reaction mixture was diluted with water, extracted with diethyl ether (3 times). The combined organic phases were washed with water, saturated $NaHCO_3$ and brine, dried ($MgSO_4$) and concentrated. The crude product was purified by flash chromatography on silica gel, eluting with 0-30% EtOAc/Hexane to afford **17** (20.9 g, 92%). ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.76 – 7.57 (m, 4H), 7.50 – 7.31 (m, 6H), 5.04 (t, *J* = 9.5 Hz, 1H), 4.19 (dd, *J* = 11.1, 1.6 Hz, 1H), 3.69 (dd, *J* = 7.8, 0.9 Hz, 1H), 2.83 (dd, *J* = 11.2, 1.1 Hz, 1H), 2.46 – 2.14 (m, 3H), 2.14 – 1.97 (m, 1H), 1.93 – 1.74 (m, 3H), 1.45 (s, 9H), 1.08 (s, 9H), 0.80 (ddt, *J* = 24.2, 11.4, 5.9 Hz, 2H), 0.57 – 0.42 (m, 1H), 0.36 (dt, *J* = 8.8, 5.2 Hz, 1H). Mass Calcd: 565.29. m/z found: 566.3.

4.4.3. Synthesis of (3S,4aR)-3-(1-((tert-butyldiphenylsilyloxy)methyl) cyclopropyl) hexahydrocyclopenta[b][1,4]oxazin-2(3H)-one **18**

To compound **17** (18.46 g, 32.6 mmol) in CH_2Cl_2 (210 mL) was added TFA (60 mL). The reaction was stirred for 2 h. TLC analysis (20% EtOAc/Hexane) showed that the reaction was complete. The reaction was diluted with saturated $NaHCO_3$ at 0 ºC slowly and extracted with CH_2Cl_2 (3 times). The combined organic phases were washed with water, brine, dried ($MgSO_4$)

and concentrated. The crude product was purified by flash chromatography on silica gel, eluting with 0 to 40% EtOAc/Hexane to afford cyclized imine intermediate (12.7 g, 87.0% total yield).

To a resulting cyclized imine product (12.7 g, 28.4 mmol) in CH_2Cl_2 (120 mL), was added $NaBH(OAc)_3$ (18.0 g, 85 mmol) and HOAc (0.5 mL) at RT. Then the reaction was stirred at 45 ºC overnight. TLC analysis (4% MeOH/DCM) showed that the reaction was complete. The reaction cooled down to 0 ºC, quenched with saturated $NaHCO_3$, extracted with CH_2Cl_2 (3 times). The combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated. The crude product was purified by flash chromatography, eluting with 0 – 45% EtOAc/Hexane to afford compound **18** (9.15g, 75%). ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.74 – 7.56 (m, 4H), 7.48 – 7.29 (m, 6H), 4.57 (q, *J* = 6.2 Hz, 1H), 4.22 – 4.09 (m, 1H), 3.42 (q, *J* = 6.1 Hz, 1H), 3.23 (d, *J* = 10.8 Hz, 1H), 2.74 (d, *J* = 8.8 Hz, 1H), 2.49 – 2.32 (m, 1H), 2.14 – 1.89 (m, 3H), 1.89 – 1.70 (m, 1H), 1.61 – 1.41 (m, 2H), 0.70 – 0.45 (m, 4H). ^{13}C NMR (Chloroform-*d*) δ (ppm): 171.50, 135.63, 135.59, 133.19, 133.14, 129.74, 129.72, 127.69, 127.68, 81.00, 77.26, 76.45, 68.35, 62.57, 55.71, 31.98, 31.89, 26.87, 22.96, 20.92, 19.18, 10.40, 9.50. Mass Calcd: 449.24. m/z found: 450.1.

4.4.4. Synthesis of (3S,4aR,7aS)-3-(1-((tert-butyldiphenylsilyloxy)methyl)cyclopropyl)-4-(4-chlorophenylsulfonyl) hexahydrocyclopenta[b][1,4]oxazin-2(3H)-one **19a** and (3S,4aR,7aR)-3-(1-(((tert-butyldiphenylsilyl)oxy)methyl)cyclopropyl)-4-((4-chlorophenyl)sulfonyl)hexahydrocyclopenta[b][1,4]oxazin-2(3H)-one **19b**

To a solution of **18** (4.95 g, 11.0 mmol) in pyridine (100.0 mL) in a sealed tube, was added 4-chlorobenzene-1-sulfonyl chloride (7.89 g, 37.4 mmol or 9.29 g, 44.0 mmol) as solid. Then the reaction was stirred at RT overnight. TLC analysis (both pure CH_2Cl_2 and 25% EtOAc/Hexane) showed that the reaction was complete. The reaction was diluted with water, extracted with CH_2Cl_2 (3 times). The combined organic phases were washed with saturated $NaHCO_3$, water and brine, dried ($MgSO_4$) and concentrated. The residue was purification by flash chromatography,

eluting with 0 to 30% EtOAc/Hexane to afford **19a** (5.0 g, 73%). ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.79 – 7.55 (m, 6H), 7.55 – 7.30 (m, 8H), 4.42 (s, 1H), 4.26 (td, *J* = 4.6, 2.2 Hz, 1H), 4.11 (ddd, *J* = 11.3, 7.9, 4.8 Hz, 1H), 3.79 (d, *J* = 10.7 Hz, 1H), 3.29 (d, *J* = 10.7 Hz, 1H), 2.11 – 1.94 (m, 1H), 1.88 (dq, *J* = 9.0, 3.9, 2.0 Hz, 2H), 1.81 – 1.63 (m, 1H), 1.63 – 1.41 (m, 1H), 1.26 (td, *J* = 7.1, 0.7 Hz, 1H), 0.98 – 0.85 (m, 1H), 0.82 (dt, *J* = 9.7, 5.8 Hz, 1H), 0.69 (dt, *J* = 9.7, 5.8 Hz, 1H), 0.46 (ddd, *J* = 9.6, 6.1, 4.7 Hz, 1H). ^{13}C NMR (Chloroform-*d*) δ (ppm): 165.50, 139.80, 138.02, 135.86, 135.67, 133.35, 133.12, 129.78, 129.69, 129.67, 128.46, 127.64, 127.62, 80.52, 77.26, 76.75, 65.59, 56.92, 54.41, 30.64, 26.79, 26.74, 24.68, 19.78, 19.20, 10.88, 9.42. Mass Calcd: 623.19. m/z found: 624.3. Refer to analysis by 2D NMR in the supporting information.

Compound **19b** was prepared analogously to compound **19a**, but starting from a diastereomeric mixture of starting material, and separating compound **19a** by column chromatography (silica gel, 25% EtOAc/Hexane)**:** ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.81 – 7.66 (m, 2H), 7.66 – 7.56 (m, 4H), 7.56 – 7.46 (m, 2H), 7.46 – 7.31 (m, 6H), 4.97 (td, *J* = 10.4, 7.5 Hz, 1H), 4.46 (s, 1H), 3.82 – 3.61 (m, 1H), 3.41 (d, *J* = 11.4 Hz, 1H), 2.87 (td, *J* = 10.5, 7.1 Hz, 1H), 2.44 – 2.20 (m, 1H), 2.06 – 1.61 (m, 5H), 1.19 – 0.84 (m, 11H), 0.50 (m, 2H). ^{13}C NMR (Chloroform-*d*) δ (ppm): 168.36, 140.43, 135.83, 135.61, 134.40, 133.21, 132.59, 129.86, 129.81, 129.25, 127.73, 127.69, 77.55, 77.25, 77.00, 67.29, 62.15, 60.56, 27.83, 26.66, 25.80, 25.26, 19.21, 18.09, 11.14, 8.31. Mass Calcd: 623.19. m/z found: 624.3. Refer to analysis by 2D NMR in the supporting information.

4.4.5. Synthesis of (3S,4aR,7aS)-3-(1-((tert-butyldiphenylsilyloxy)methyl)cyclopropyl)-4-(4-chlorophenylsulfonyl) octahydrocyclopenta[b][1,4]oxazine **20**

To compound **19a** (5.0 g, 8.01 mmol) and $CaCl_2$ (5.33 g, 48.1 mmol) in THF (75.0 mL) and EtOH (75.0ml) was added $NaBH_4$ (1.51 g, 40.0 mmol) as solid. Then the reaction was stirred at

RT overnight. TLC analysis (30% EtOAc/Hexane) showed that the reaction was complete. The reaction was quenched with citric acid (20%) at 0 ºC, extracted with CH_2Cl_2 (3 times). The combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated. The residue was purified by flash chromatography on silica gel, eluting with 0 to 30% EtOAc/Hexane to afford the diol product (3.1g, 62%).

To the diol starting material (3.12 g, 4.97 mmol) obtained above and PPh_3 (2.60 g, 9.93 mmol) in THF (45.0 mL) was added DEAD (1.57 mL, 9.93 mmol) at RT. The reaction was stirred at RT overnight. TLC analysis (25% EtOAc/Hexane) showed that the reaction was complete with formation of a less polar product. The reaction mixture was diluted with water, extracted with CH_2Cl_2 (3 times). The combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated. The crude product was purified by flash chromatography on silica gel, eluting with 0 to 35% EtOAc/Hexane to afford the product **20** (2.43 g, 80%). ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.75 – 7.55 (m, 6H), 7.48 – 7.28 (m, 8H), 4.00 (d, *J* = 4.3 Hz, 1H), 3.86 (d, *J* = 10.3 Hz, 1H), 3.78 – 3.54 (m, 3H), 3.32 (t, *J* = 5.0 Hz, 1H), 3.12 (dd, *J* = 12.4, 4.3 Hz, 1H), 1.89 – 1.47 (m, 5H), 1.45 – 1.21 (m, 1H), 1.05 (s, 9H), 0.99 – 0.79 (m, 1H), 0.78 – 0.64 (m, 2H), 0.63 – 0.48 (m, 1H). Mass Calcd: 609.21. m/z found: 610.3.

4.4.6. Synthesis of (1-((3S,4aR,7aS)-4-((4-chlorophenyl)sulfonyl) octahydrocyclopenta[b][1,4]oxazin-3-yl)cyclopropyl)methanol **21**

To a starting compound **20** (970 mg, 1.59 mmol) in THF (15 mL) was added TBAF (1 M in THF, 3.2 mmol) at RT. The reaction was stirred at RT for 2 h. TLC analysis (80% CH_2Cl_2/Hexane) showed that the reaction was complete. The reaction mixture was quenched with water, extracted with EtOAc (3 times). The combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated. The crude was purified by flash chromatography on silica gel, eluting with 0 to 30% EtOAc/Hexane to obtain desired product **21**

(450 mg, 76.0%). Compound **21** was further resolved by Chiral OJ column to obtain a single enantiomer. $[\alpha]_D^{20}$ = -123.71 (EtOH). ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.84 – 7.62 (m, 2H), 7.61 – 7.38 (m, 2H), 4.35 (ddd, *J* = 12.5, 4.9, 2.3 Hz, 1H), 4.04 (d, *J* = 4.1 Hz, 1H), 3.87 (ddd, *J* = 10.5, 8.6, 4.3 Hz, 1H), 3.62 (dd, *J* = 10.0, 4.8 Hz, 1H), 3.36 (d, *J* = 12.8 Hz, 1H), 3.23 (t, *J* = 4.6 Hz, 1H), 2.83 (dd, *J* = 12.8, 4.2 Hz, 1H), 2.71 (dd, *J* = 12.5, 9.9 Hz, 1H), 2.19 – 1.89 (m, 2H), 1.78 (tdd, *J* = 11.5, 7.7, 5.6 Hz, 2H), 1.70 – 1.55 (m, 1H), 1.55 – 1.37 (m, 1H), 1.21 (dt, *J* = 9.3, 5.6 Hz, 1H), 0.74 (dt, *J* = 9.5, 5.8 Hz, 1H), 0.61 (dtd, *J* = 7.8, 5.4, 2.2 Hz, 1H), 0.50 (dt, *J* = 10.0, 5.3 Hz, 1H). Mass Calcd: 371.1. m/z found: 372.3.

4.4.7. Synthesis of (3S,4aR,7aS)-4-((4-chlorophenyl)sulfonyl)-3-(1-(iodomethyl)cyclopropyl) octahydrocyclopenta[b][1,4]oxazine **22**

The starting hydroxyl compound **21** (860 mg, 2.31 mmol) in CH_3CN (4 mL) and toluene (8 mL) was treated with Ph_3P (728 mg, 2.78 mmol), I_2 (704 mg, 2.78 mmol) and imidazole (472 mg, 6.94 mmol) at RT. The reaction was stirred at RT for 3 h. TLC analysis (30% EtOAc/Hexane) showed that the reaction was complete. The reaction mixture was quenched with NH_4Cl, extracted with EtOEt (3 times). The combined organic phases were washed with saturated $NaHCO_3$ and brine, dried ($MgSO_4$) and concentrated. The residue was purified by flash chromatography on silica gel, eluting with 0 to 80% CH_2Cl_2/Hexane in 1h to afford the desired product **22** (1.0 g, 90%). ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.91 – 7.72 (m, 2H), 7.59 – 7.40 (m, 2H), 4.10 (d, *J* = 4.1 Hz, 1H), 3.91 (dd, *J* = 10.3, 1.4 Hz, 1H), 3.81 (td, *J* = 9.6, 4.7 Hz, 1H), 3.58 (d, *J* = 12.8 Hz, 1H), 3.29 (td, *J* = 4.9, 1.3 Hz, 1H), 3.12 (d, *J* = 10.3 Hz, 1H), 3.01 (dd, *J* = 12.8, 4.2 Hz, 1H), 1.99 (td, *J* = 8.9, 6.3 Hz, 2H), 1.89 – 1.69 (m, 3H), 1.69 – 1.37 (m, 2H), 1.03 – 0.82 (m, 2H), 0.69 (ddd, *J* = 8.9, 6.9, 4.8 Hz, 1H). Mass Calcd: 481.00. m/z found: 482.1.

4.4.8. Synthesis of 2-(1-((3S,4aR,7aS)-4-((4-chlorophenyl)sulfonyl) octahydrocyclopenta[b][1,4]oxazin-3-yl)cyclopropyl)acetonitrile **23**

To a suspension of the iodine compound **22** (600 mg, 1.24 mmol) in CH_3CN (12 mL) was added n-Bu_4NCN (736 mg, 2.74 mmol, not soluble) at RT. The reaction mixture was stirred at RT for 2 h. TLC analysis (70% CH_2Cl_2/Hexane) showed that the reaction was complete. The reaction diluted with water and the mixture was extracted with EtOAc (3 times). The combined organic phases were washed with water and brine, dried ($MgSO_4$) and concentrated. The crude product was purified by flash chromatography, eluting with 0 to 30% EtOAc/Hexane in 40 minutes to give compound **23** (420 mg, 89%). ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.83 – 7.65 (m, 2H), 7.59 – 7.44 (m, 2H), 3.94 (d, *J* = 4.1 Hz, 1H), 3.81 (td, *J* = 9.6, 4.3 Hz, 1H), 3.37 (d, *J* = 12.9 Hz, 1H), 3.22 – 3.11 (m, 1H), 3.06 (d, *J* = 17.4 Hz, 1H), 2.92 – 2.72 (m, 2H), 2.12 – 1.93 (m, 2H), 1.89 – 1.68 (m, 2H), 1.68 – 1.41 (m, 2H), 1.40 – 1.17 (m, 1H), 0.93 – 0.75 (m, 2H), 0.75 – 0.58 (m, 1H). Mass Calcd: 380.10. m/z found: 381.3.

4.4.9. Synthesis of 2-(1-((3S,4aR,7aS)-4-((4-chlorophenyl)sulfonyl)octahydrocyclopenta[b][1,4] oxazin-3-yl)cyclopropyl)acetic acid **24**

To the nitrile starting material **23** (420 mg, 1.103 mmol) in CH_2Cl_2 (10 mL) was added DIBAL-H (1.3 mL, 1.3 mmol) at -78 ºC dropwise. The reaction was stirred at -78 ºC for 4 h. TLC analysis (10% EtOAc/Hexane) showed that the reaction was complete. The reaction mixture was quenched with MeOH (3 mL) slowly and stirred for 10 minutes, H_2SO_4 (1N, 3 mL) was added and stirred for 45 minutes and extracted with CH_2Cl_2 (3 times). The combined organic phases were washed with water, brine, dried ($MgSO_4$) and concentrated. The crude product was purified by flash chromatography on silica gel, eluting with 0 to 20% EtOAc/Hexane to afford the aldehyde intermediate product (0.40g, 94%).

To the starting aldehyde intermediate (400 mg, 0.85mmol) obtained above in t-BuOH (14 mL) and water (3 mL) at 0 ºC, was added 2-methyl-2-butene (1.7 ml, 2M in THF, 3.4 mmol), followed by sodium chlorite (472 mg, 5.22 mmol). The reaction was stirred at RT for 2 h. TLC

analysis (30% EtOAc/Hexane) showed that the reaction was complete. The reaction was quenched by saturated NH_4Cl (10 mL) and extracted with EtOAc (3×25 mL). The combined organic phases were washed with water, brine, dried ($MgSO_4$) and concentrated. The crude product was purified by flash chromatography on silica gel, eluting with 0 to 30% EtOAc/Hexane to afford **24** (230 mg, 55 %). : ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.83 – 7.61 (m, 2H), 7.53 – 7.36 (m, 2H), 4.19 – 3.98 (m, 1H), 3.92 – 3.73 (m, 1H), 3.48 – 3.07 (m, 3H), 2.78 (dd, *J* = 12.9, 4.2 Hz, 1H), 2.16 – 1.84 (m, 3H), 1.75 (tdd, *J* = 13.4, 10.6, 6.5 Hz, 2H), 1.65 – 1.52 (m, 1H), 1.52 – 1.34 (m, 1H), 1.24 (m, 1H), 0.86 – 0.71 (m, 1H), 0.63 (dt, *J* = 9.2, 6.2 Hz, 1H), 0.48 (dt, *J* = 10.1, 5.4 Hz, 1H). Mass Calcd: 399.09. m/z found: 400. 3.

4.4.10. General procedure for synthesis of final compounds **25a-e**

Compounds **25a-e** were prepared by using the general procedure for synthesis of final compounds **15a-i**.

Compound **25a**. Yield: 84%. ^{1}H NMR: 7.74 (2H, d, J = 8.67), 7.46 (2H, d, J = 8.60), 4.70 (0.5H, d, J = 13.33), 4.63 (0.5H, d, J = 12.94), 4.31 (0.5H, d, J = 3.88), 4.21 (0.5H, d, J = 3.72), 3.76 – 3.88 (2H, m), 3.36 – 3.50 (4H, m), 3.12 (1H, m), 3.06 (0.5H, t, J = 13.07), 2.95 (0.5H, t, J = 12.20), 2.79 (1H, dd, J = 12.82, 4.20), 2.61 (0.5H, t, J = 13.53), 2.52 (0.5H, t, J = 12.65), 2.17 (1H, m), 2.03 (1H, m), 1.30 – 1.84 (10H, m), 1.24 (1H, m), 1.13 (1H, m), 0.86 (1H, m), 0.63 (1H, m), 0.42 (1H, m). Mass Calcd: 496.18. m/z found: 497.3.

Compound **25b**. Yield: 74%. ^{1}H NMR: 7.74 (2H, dd, J = 6.36, 8.39), 7.46 (2H, dd, J = 5.74, 8.49), 4.73 (0.5H, d, J = 13.46), 4.65 (0.5H, d, J = 13.10), 4.30 (0.5H, d, J = 3.95), 4.19 (0.5H, d, J = 3.78), 3.77 – 3.90 (2H, m), 3.44 (1H, d, J = 12.83), 3.38 (1H, dd, J = 4.78, 16.49), 3.13 (1H, m), 3.04 (0.5H, t, J = 12.76), 2.91 (0.5 H, m), 2.78 (1H, dd, J = 4.11, 12.92), 2.48 (6H, s), 2.16

(1H, m), 2.03 (1H, m), 1.53 – 1.83 (13H, m), 1.40 (2H, m), 1.24 (1H, m), 0.85 (1H, m), 0.62 (1H, m), 0.42 (1H, m). Mass Calcd: 549.24. m/z found: 550.3.

Compound **25c**. Yield: 69 %. ^{1}H NMR: 7.73 (2H, d, J = 8.49), 7.47 (2H d, J = 8.51), 4.23 (1H, dd, J = 3.92, 10.65), 3.95 (1H, m), 3.79 (2H, m), 3.61 (2H, m), 3.46 (4H, m), 3.33 (2H, m), 3.13 (1H, q, J = 5.10), 2.78 (1H, dt, J = 12.87, 3.94), 2.01 – 2.19 (2H, m), 2.09 (3H, s), 1.49 – 1.82 (5H, m), 1.28 (1H, m), 0.90 (1H, m), 0.65 (1H, m), 0.42 (1H, m). Mass Calcd: 509.18. m/z found: 510.3.

Compound **25d**. Yield: 82%. ^{1}H NMR: 7.73 (2H, m), 7.47 (2H, dd, J = 1.54, 8.33), 4.32 – 4.56 (1H, m), 4.18 (1H, m), 3.20 – 3.93 (7H, m), 3.13 (1H, m), 2.61 – 2.80 (2H, m), 1.48 – 2.20 (9H, m), 1.24 (1H, m), 0.70 – 0.91 (2H, m), 0.42 (1H, m). Mass Calcd: 468.15. m/z found: 469.3.

Compound **25e**. Yield: 83 %. ^{1}H NMR: 7.73 (2H, d, J = 8.62), 7.47 (2H, d, J = 8.23), 4.56 (0.5 H, s), 4.34 (0.5 H, s), 4.22 (0.5 H, d, J = 3.88), 4.14 (0.5 H, d, J = 4.25), 4.12 (0.5 H, J not measuable), 4.00 (0.5 H, d, J = 11.55), 3.81 – 3.94 (1H, m), 3.64 (1H, m), 3.53 (1H, m), 3.39 (3H, m), 3.17 (1H, dt, J = 22.15, 4.57), 2.72 (1H, td, J = 13.77, 4.13), 2.17 (1H, m), 2.02 (3H, m), 1.51 – 1.93 (8H, m), 1.26 (1H, m), 0.89 (1H, m), 0.71 (1H, m), 0.42 (1H, m). Mass Calcd: 482.16. m/z found: 483.3.

4.5. Procedures for Synthesis of alpha-hydroxy amide morpholine compounds **29a-b**

4.5.1. Synthesis of 2-((tert-butyldimethylsilyl)oxy)-2-(1-((3S,5R)-4-((4-chlorophenyl)sulfonyl)-5-cyclopropylmorpholin-3-yl)cyclopropyl)acetonitrile **26**

To compound **11** (1.36 g, 3.66 mmol) in CH_2Cl_2 (50 mL) was added Dess-Martin periodinane (3.1 g, 7.31 mmol) as solid. The reaction was stirred for 2 h. TLC analysis (30% EtOAc/hexane) showed that the reaction was completed with formation of a less polar product. The reaction was quenched with saturated $NaHCO_3$ (15 mL) and $Na_2S_2O_3$ (600 mg) and stirred for 1h. The

mixture was extracted with CH_2Cl_2 (3 times). The combined organic phases were washed with brine, dried ($MgSO_4$) and concentrated to obtain crude product. The crude product was purified by flash chromatography on silica gel, eluting with 0 - 30% EtOAc/Hexane to obtain aldehyde product (800 mg, 59%).

To the aldehyde (800 mg, 2.16 mmol) in CH_3CN (15 mL), was added KCN (843 mg, 12.98 mmol), TBSCl (488 mg, 3.24 mmol) and ZnI_2 (34.45 mg, 0.108 mmol). The reaction mixture was stirred at 55 ºC overnight. The reaction was quenched with water, extracted with CH_2Cl_2 (3 times), dried and concentrated. The crude product was purified by flash chromatography, eluting with 0 - 30% EtOAc/Hexane to give diastereomer A 390 mg (35%, not pure), and B (compound **26**, 563 mg, 51%). ^{1}H NMR: 7.78 (2H, d, J = 8.82), 7.51 (2H, d, J = 8.81), 5.02 (1H, s), 4.14 (1H, d, J = 4.38), 3.75 (1H, d, J = 11.52), 3.51 (1H, d, J = 12.73), 3.08 (1H, d, J = 4.04, 11.52), 2.90 (1H, dd, J = 4.59, 12.75), 2.84 (1H, dd, J = 3.91, 10.26), 1.55 (1H, m), 1.37 (1H, m), 1.17 (1H, m), 0.89 (9H, s), 0.89 (1H, m), 0.71 (1H, m), 0.62 (2H, m), 0.53 (1H, m), 0.26 (3H, s), 0.26 (1H, m), 0.16 (3H, s). Mass Calcd: 510.18. m/z found: 511.4.

4.5.2. Synthesis of 2-(tert-butyldimethylsilyloxy)-2-(1-((3S,5R)-4-(4-chlorophenylsulfonyl)-5-cyclopropylmorpholin-3-yl)cyclopropyl)acetaldehyde **27**

To compound **26** (560 mg, 0.76 mmol) in CH_2Cl_2 (10 mL) at -78 ºC was added DIBAL-H (1 M in hexane, 1.38 mL, 1.38 mmol). The mixture was stirred for 4 h and quenched with 20% sodium tartrate buffer (pH 4) and extracted with CH_2Cl_2 (3 times). The combined organic phases were washed with brine, dried ($MgSO_4$) and concentrated. The crude product was prified by flash chromatography, eluting with 0-30% EtOAc/Hexane to obtain product **27** (198 mg, 35%). ^{1}H NMR: 9.81 (1H, s), 7.68 (2H, d, J = 8.77), 7.48 (2H, d, J = 8.77), 4.26 (1H, s), 4.00 (1H, d, J = 4.66), 3.73 (1H, d, J = 11.42), 3.66 (1H, d, J = 12.47), 3.04 (1H, dd, J = 4.24, 11.38), 2.97 (1H, dd, J = 5.03, 12.46), 2.76 (1H, dd, J = 3.93, 10.26), 1.45 (1H, m), 1.32 (1H, m), 0.95 (1H, m),

0.89 (9H, s), 0.56 – 0.73 (5H, m), 0.22 (1H, m), 0.14 (3H, s), 0.05 (3H, s). Mass Calcd: 513.18. m/z found: 514.3.

4.5.3. Synthesis of 2-(1-((3S,5R)-4-(4-chlorophenylsulfonyl)-5-cyclopropylmorpholin-3-yl)cyclopropyl)-2-hydroxyacetic acid **28**

To the aldehyde compound **27** (140 mg, 0.27 mmol) in a mixture of t-BuOH/water (2 mL/0.5 mL) and 2-methyl-2butene (2M in THF, 0.87 mL, 1.74 mmol) was added $NaH_2PO_4 \cdot H_2O$ (75 mg, 0.544 mmol) and $NaClO_2$ (78.71 mg, 0.87 mmol). The mixture was stirred overnight. The mixture was partitioned between CH_2Cl_2 and 20% citric acid, re-extracted the aqueous phase with CH_2Cl_2 (3 times). The combined organic phases were washed with brine, dried ($MgSO_4$) and concentrated (125 mg, 87.0%).

To the carboxylic acid obtained above (110 mg, 0.21 mmol), Diastereomeric series B, in THF (3 mL) was added TBAF (1M in THF, 0.72 mmol, 0.72 mL) at RT. The mixture was stirred for 3 h. LCMS analysis indicated that the reaction was complete. The reaction was quenched with water, extracted with CH_2Cl_2 (3 times). The combined organic layers were washed with water and brine, dried ($MgSO_4$) and concentrated. The residue was purified by Gilson reverse phase HPLC to obtain compound **28** (61 mg, 71%). ^{1}H NMR: 7.74 (2H, d, J = 8.82), 7.53 (2H, d, J = 8.56), 4.79 (1H, s), 4.08 (1H, d, J = 3.94), 3.76 (1H, d, J = 11.51), 3.50 (1H, d, J = 12.99), 3.02 (1H, dd, J = 3.96, 11.54), 2.92 (1H, dd, J = 4.56, 12.74), 2.83 (1H, dd, J = 3.85, 10.30), 1.65 (1H, m), 1.25 – 1.37 (2H, m), 0.61 – 0.77 (5H, m), 0.29 (1H, m). Mass Calcd: 415.09. m/z found: 416.3.

4.5.4. Synthesis of 1-(3,8-diazabicyclo[3.2.1]octan-3-yl)-2-(1-((3S,5R)-4-(4-chlorophenylsulfonyl)-5-cyclopropylmorpholin-3-yl)cyclopropyl)-2-hydroxyethanone **29a**

To the carboxylic acid **28** (60 mg, 0.144 mmol) and tert-butyl (1R,5S)-3,8-diazabicyclo[3.2.1]octane-8-carboxylate (61mg, 0.29mmol) in CH_2Cl_2 (1.0 mL) was added BOP

(65 mg, 0.144 mmol), followed by NMM (47 µl, 0.43 mmol). The mixture was stirred at RT for 5 h. LCMS analysis showed that the reaction was done. The reaction was quenched with brine, extracted with EtOAc (3 times) and CH_2Cl_2 (2 times). The organic layers were washed with water and brine, dried ($MgSO_4$) and concentrated. The residue was purified by prepare TLC (4% MeOH/ CH_2Cl_2) to obtain one major fraction after purification (60 mg, 68 %).

To the Boc-protected amine product in CH_2Cl_2 (5 mL) was added TFA (1 mL). The mixture was stirred for 50 minutes. TLC analysis (10% MeOH/ CH_2Cl_2) showed that the reaction was done. Solvent was removed. The residue was dissolved in CH_2Cl_2, washed with NaOH (2 M), water and brine, dried ($MgSO_4$) and concentrated. The crude product was purified by prepare TLC (10% MeOH/CH_2Cl_2) to afford **29a** (37.9 mg, 76%). ^{1}H NMR: 7.72 (2H, m), 7.47 (2H, d, J = 8.42), 5.289 (0.4H, d, J = 6.62), 5.14 (0.6 H, d, J = 6.62), 4.17 (1H, d, J = 12.93), 3.89 – 4.06 (2H, m), 3.46 – 3.76 (5H, m), 3.35 (3H,, m), 3.15 (1H, m), 2.72 – 3.0 (2H, m), 1.53 – 1.85 (5H, m), 1.08 (1H, m), 0.66 – 0.83 (5H, m), 0.47 (1H, m), 0.26 (1H, m). Mass Calcd: 509.18. m/z found: 510.4.

4.5.5. Synthesis of 2-(1-((3S,5R)-4-(4-chlorophenylsulfonyl)-5-cyclopropylmorpholin-3-yl)cyclopropyl)-2-hydroxy-1-(3-(piperidin-1-yl)-8-azabicyclo[3.2.1]octan-8-yl)ethanone **29b**

To the 3-(piperidin-1-yl)-8-azabicyclo[3.2.1]octane hydrochloride (104 mg, 0.45 mmol) in CH_2Cl_2 (2.0 mL) was added Et_3N (64 µl, 0.453 mmol). Followed by the starting acid **28** (62.5 mg, 0.15 mmol) in CH_2Cl_2 (1 mL), BOP (67 mg, 0.15 mmol) and NMM (50 µl, 0.453 mmol). The reaction was stirred at RT overnight. The reaction mixture was quenched with brine, extracted with CH_2Cl_2 (3 times). The combined organic layers were washed with water and brine, dried ($MgSO_4$) and concentrated. The crude product was purified by prepare TLC (10% MeOH/CH_2Cl_2) to obtain a major product **29b** after purification (34 mg, 38.2%). ^{1}H NMR: 7.72 (2H, d, J = 8.51), 7.47 (2H, d, J = 8.48), 5.05 (1H, d, J = 8.24), 4.69 (1H, d, J = 7.46), 4.55 (1H,

d, J = 7.91), 4.08 (1H, dd, J = 7.42, 12.28), 3.71 (1H, d, J = 10.71), 3.30 – 3.50 (4H, m), 2.86 (1H, t, J = 9.03), 2.76 (1H, m), 2.44 (4H, bs), 1.53 – 2.00 (14H, m), 1.42 (2H, m), 1.01 (1H, m), 0.56 – 0.81 (5H, m), 0.26 (1H, m). Mass Calcd: 591.25. m/z found: 592.3.

AUTHOR INFORMATION

Corresponding Author

*Phone: 908-740-2383. E-mail: zhiqiang.zhao@merck.com

ACKNOWLEDGMENT

We Thank Dr. Alexander Pasternak for his support and review of this paper, and Analytical Chemistry group for LCMS analysis.

References

[1] 2015 Alzheimer's disease facts and figures, http://www.alz.org/

[2] Hardy, J.; Selkoe, D. *J.* The amyloid hypothesis of Alzheimer's disease: progress and problems on the road to therapeutics. *Science* **2002**, 297, 353-356.

[3] (a) Josien, H. Recent advances in the development of gamma-secretase inhibitors. *Opin. Drug Discov.* **2002**, *5*, 513-525; (b) Ziani-Cherif, C.; Mostefa-Kara, B.; Brixi-Gormat, F. Z. Gamma-secretase as a pharmacological target in Alzheimer disease research: when, why and how? *Curr. Pharm. Design* **2006**, *12*, 4313-4335; (c) Evin, G.; Sernee, M. F.; Masters, C. L. Inhibition of gamma-secretase as a therapeutic intervention for Alzheimer's disease: prospects, limitations and strategies. *CNS Drugs.* **2006**, *20*, 351-372.

[4] Wu, W-L.; Zhang, L. γ-Secretase inhibitors for the treatment of Alzheimer's disease. *Drug Dev. Res.* **2009**, *70*, 94-100.

[5] Kreft, A. F.; Martone, R.; and Porte, A. Recent advances in the identification of γ-secretase inhibitors to clinically test the Aβ oligomer hypothesis of Alzheimer's disease. *J. Med. Chem*. **2009**, *52*, 6169–6188

[6] Deangelo, D. J.; Stone, R. M.; Silverman, L. B.; Stock, W.; Attar, E. C.; Fearen, I.; Dallob, A.; Matthews, C.; Stone, J.; Freedman, S. J.; Aster, J. A phase I clinical trial of the Notch inhibitor MK-0752 in patients with T-cell acute lymphoblastic leukemia/lymphoma (T-ALL) and other leukemias. *J. Clin. Oncol*. **2006**, *24*, 6585.

[7] Schott, A. F.; Landis, M. D.; Dontu, G.; Griffith, K. A.; Layman, R. M.; Krop, I.; Paskett, L. A.; Wong, H.; Dobrolecki, L. E.; Lewis, M. T.; Froehlich, A. M.; Paranilam, J.; Hayes, D. F.; Wicha, M. S.; Chang, J. C. Preclinical and clinical studies of gamma secretase inhibitors with docetaxel on human breast tumors. *Clin. Cancer Res*. **2013**, *19*, 1512-1524.

[8] Josien, H.; Bara, T.; Rajagopalan, M.; Asberom, T.; Clader, J. W.; Favreau, L.; Greenlee, W. J.; Hyde, L. A.; Nomeir, A. A.; Parker, E. M.; Pissarnitski, D. A.; Song, L.; Wong, W. T.; Zhang, L.; Zhang, Q. and Zhao, Z. Small conformationally restricted piperidine N-arylsulfonamides as orally active γ-secretase inhibitors, *Bioorg. Med. Chem. Lett*., **2007**, *17*, 5330-5335.

[9] Asberom, T.; Zhao, Z.; Bara, T.; Clader, J. W.; Greenlee, W. J.; Hyde, L. A.; Josien, H. B.; Li, W.; McPhail, A. T.; Nomeir, A. A.; Parker, E. M.; Rajagopalan, M.; Song, L.; Wong, G. T.; Zhang, L.; Zhang, Q. and Pissarnitski, D. A. Discovery of γ-secretase inhibitors efficacious in a transgenic aninal model of Alzheimer's disease, *Bioorg. Med. Chem. Lett*. **2007**, 17, 511-516.

[10] Li, H.; Asberom, T; Bara, T.; Clader, J.; Greenlee, W. J.; Josien, H. B.; McBriar, M. D.; Nomeir A.; Pissarnitski, D. A.; Rajagopalan, M.; Xu, R..; Zhao, Z.; Song, L.; Zhang, L. Discovery of 2,4,6-trisubstituted N-arylsulfonyl piperidines as γ-secretase inhibitors, *Bioorg. Med. Chem. Lett.* **2007**, *17*, 6290-6294.

[11] Josien H.; Clader, J. W.; Bara, T. A.; Xu, R.; Li, H.; Pissarnitski, D.; Zhao, Z. Preparation of substituted N-arylsulfonylheterocyclic amines as γ-secretase inhibitors. PCT Int. *Appl.* **2008**, WO 2014085211 A2 20140605.

[12] Chang, Xiao-Wei; Han, Qing-Chuan; Jiao, Zhi-Gang; Weng, Lin-Hong; Zhang, Dan-Wei. 1-Aminoxymethylcyclopropanecarboxylic acid as building block of β N-O turn and helix: synthesis and conformational analysis in solution and in the solid state, *Tetrahedron*, **2010**, 66 (51), 9733-9737;.

[13] Kim, Y. K.; Park, S. Y.; Joo, H. W.; Choi, E. S.. Biaryl derivatives as GPR120 agonists PCT Int. Appl., WO2014209034.

[14] Please see supporting information for detailed 2D NMR analysis.

[15] Teall, M.; Oakley, P.; Harrison, T, Shaw, D.; Kay, E.; Elliott, J.; Gerhard, U.; Castro, J. L.; Shearman, M.; Ball, R. G.; Tsou, N. N. Aryl sulfones: a new class of γ-secretase inhibitors, *Bioorg. Med. Chem. Lett*. **2005**, *15*, 2685-2688.

[16**]** Churcher, I.; Beher, D.; Best, J. D.; Castro, J. L.; Clarke, E. E.; Gentry, A.; Harrison, T.; Hitzel, L.; Kay, E.; Kerrad, S.; Lewis, H. D.; Morentin-Gutierrez, P.; Mortishire-Smith, R.; Oakley, P. J.; Reilly, M.; Shaw, D. E.; Shearman, M. S.; Teall, M. R.; Williams, S.; Wrigley, J. D. J. 4-Substituted cyclohexyl sulfones as potent, orally active γ-secretase inhibitors, *Bioorg. Med. Chem. Lett.* **2006**, *16*, 280-284.

[17] Zhao, Z. Pissarnitski, D.; Josien, H.; Wu, W-L.; Xu, R.; Li, H.; Clader, J. W.; Burnett, D. A.; Terracina, G.; Hyde, L.; Lee, J.; Song, L.; Zhang, L.; Parker, E. M. Discovery of novel, potent γ-secretase inhibitors, *J. Med. Chem.* **2015**, 58, 8806-8817.

[18] Please see HPLC spectra of compound purity in the supporting information.

[19] Zhang, L.; Song, L.; Terracina, G.; Liu, Y.; Pramanik, B. and Parker, E. Biochemical characterization of the γ-secretase activity that produces β-amyloid peptides, *Biochemistry*, **2001**, 40, 5049–5055.

[20] Chishti, M. A.; Yang, D. S.; Janus, C. et al., Early-onset amyloid deposition and cognitive deficits in transgenic mice expressing a double mutant form of amyloid precursor protein 695, *J. Biol. Chem.*, **2001**, 276, 21562–21570.

[21]Hyde, L. A.; Kazdoba, T. M.; Grilli, M. et al., Age-progressing cognitive impairments and neuropathology in transgenic CRND8 mice, *Behavioural Brain Research*, 2005, 160 (2), 344–355.

[22] Korfmacher, W. A.; Cox, K. A.; Ng, K. J.; Veals, J.; Hsieh, Y.; Wainhaus, S.; Broske, L.; Prelusky, D.; Nomeir, A.; White, R. E. Cassette-accelerated rapid rat screen: a systematic procedure for the dosing and liquid chromatography/atmospheric pressure ionization tandem mass spectrometric analysis of new chemical entities as part of new drug discovery, *Rapid Commun. Mass Spectrom*. **2001**, 15, 335-340.

Highlight

- γ-Secretase inhibitors arrest formation of amyloid-β, which plays a role in Alzheimer's disease
- Scissile carbamate bond of a known inhibitor was replaced with the non-scissile carbon-carbon bond
- The effect of introduction of polarity (oxygen atoms) in various parts of molecule has been studied
- Stability and potency of the series was improved, while CYP3A4 inhibition was mitigated