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Total Synthesis of (−)-Balanol, All Stereoisomers, Their *N*-Tosyl Analogues, and Fully Protected Ophiocordin: An Easy Route to Hexahydroazepine Cores from Garner Aldehydes

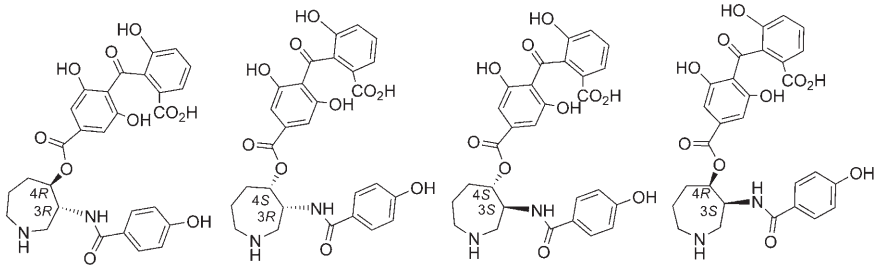
Ajay Kumar Srivastava and Gautam Panda*[a]

Abstract: Total syntheses of (−)-balanol and all of its stereoisomers starting from easily available Garner aldehydes are described. Diastereoselective Grignard reactions on Garner aldehydes and ring-closing metatheses are the key steps for the construction of hexahydroazepine subunits. The benzophenone subunits were constructed through coupling of suitably functionalized aromatic aldehyde and bromo components. The synthetic route constitutes a convenient and scalable reaction sequence to generate all of the stereoisomers of balanol. The methodology is explored further for the synthesis of *N*-tosyl analogues of balanol and of fully protected ophiocordin.

Keywords: balanol · metathesis · natural products · Protein kinase C inhibitors · total synthesis

Introduction

Protein kinase C (PKC) is a Ca^{2+}- and phospholipid-dependent enzyme that phosphorylates serine and threonine residues in a wide variety of cellular proteins.[1] PKC mediates the regulation of cardiac muscle function by a variety of neurotransmitters, hormones, and extracellular signaling molecules.[2] As PKC plays an important role in signal transduction, cell proliferation, cell differentiation, and gene expression,[3] the discovery of specific inhibitors of PKC would provide potential chemotherapeutic agents[4–7] encompassing a wide spectrum of diseases: cancer, disorders of the central nervous and cardiovascular systems, diabetes, asthma, rheumatoid arthritis, and HIV infection. Some PKC inhibitors are currently in clinical trials for various types of cancer.[8–10]



Balanol (**1a**), an unusual metabolite isolated by Kulanthaivel et al.[11–13] from the fungus *Verticillium balanoides* collected from *Pinus palustris* needle litter in 1993, and later by Ohshima et al. from *Fusarium merismoids* in 1994,[12] inhibits almost all PKC isoforms in the 4–5 nanomolar range. Ophiocordin (**3**),[14,15] a regioisomer of (−)-balanol (**1a**), was isolated from *Cordyceps ophioglossoides* in 1977 and serves as an antibiotic with antifungal activity. While several efforts to achieve selectivity among PKC isoforms have been undertaken,[16–35] there is still a constant need for more selective and potent PKC inhibitors.

In our endeavors to develop some novel anticancer agents we have recently reported various benzannulated heterocycles synthesized from naturally abundant amino acids.[36] In continuation of our program for nonbenzannulated heterocycles, we became interested in achieving the synthesis of two azepane-containing natural products (−)-balanol and ophiocordin as well as their analogues.

[a] A. K. Srivastava, Dr. G. Panda
Medicinal and Process Chemistry Division
Central Drug Research Institute, Lucknow, UP, 226001 (India)
Fax: (+91) 522-2623-938
E-mail: gautam_panda@cdri.res.in

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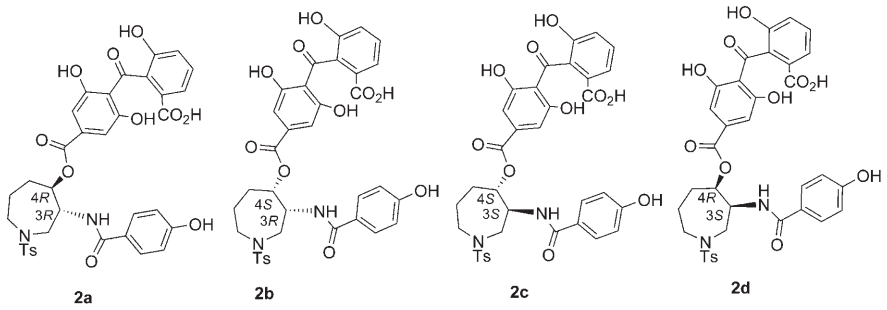
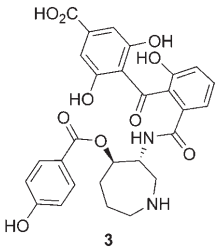


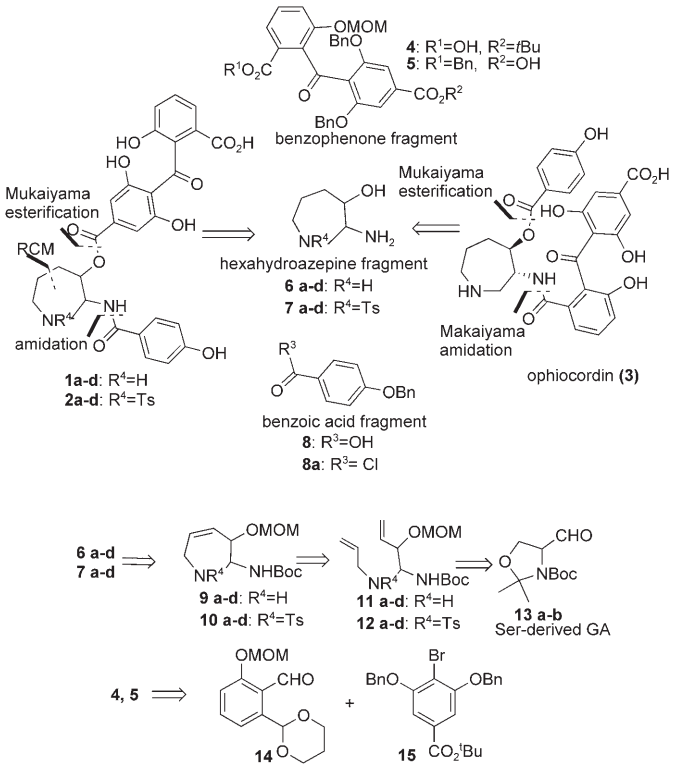
The isolation and structure elucidation of balanol (**1a**) was immediately followed by six total syntheses of the compound.[37–46] Key to Nicolaou's group's asymmetric synthesis of balanol was chiral allylation of a substituted serinal with Brown's reagent $(Ipc)_2B$-allyl.[39] An anionic homo-Fries rearrangement strategy to the benzophenone subunit and construction of the azepane unit from 3-hydroxylysine was the key step for **1a** from Lampe's and Hughes' group.[38] Tanner's group used regio- and stereoselective opening of chiral epoxides and aziridines for the synthesis of **1a**.[42] Ring expansion of piperidin-4-ones into azepane ring systems and further modification to give **1a** has also been reported.[41] A radical cyclization approach to the hexahydroazepine ring and a biomimetic route to the benzophenone fragment of **1a** have been described by Naito's group.[46] Apart from these, syntheses of the 3-amino-hexahydro-azepin-4-ol ring of balanol and suitable analogues have also been reported, because of its potent PKC inhibitory properties.[47–63] Here we report for the first time total syntheses of all stereoisomers of balanol, along with their *N*-tosyl analogues **2a–d** and also of fully protected ophiocordin, that follow a different conceptual design rooted in a flexible and diversity-oriented approach based on ring-closing metathesis (RCM)[48,51] and diastereoselective nucleophilic addition steps on Garner aldehydes.[64]



Results and Discussion

Retrosynthetic analysis and strategy: Retrosynthesis (Scheme 1) illustrates disconnection of the ester and amide linkages of **1a–d** and **2a–d** to afford the substituted hexahydroazepine units **6a–d** and **7a–d**, which can be easily accessed from their tetrahydroazepine precursors **9a–d** and **10a–d**, respectively (Scheme 1, bottom). These should in turn be obtainable through RCM[65] of the pendant vinyl groups of **11a–d** and **12a–d**, respectively. We envisioned that the divinyl derivatives **11a–d** and **12a–d** should be easily accessible from Garner aldehydes **13a–b** by diastereoselective nucleophilic addition followed by expedient functional group transformations. Similarly, the disconnection of the ester and amide linkages of ophiocordin **3** would also afford the hexahydroazepine subunit **6a** and acid **4**.

The highly congested tetra-*ortho*-substituted benzophenone fragments **4** and **5** should be preparable by coupling of



Scheme 1. Retrosynthetic analysis.

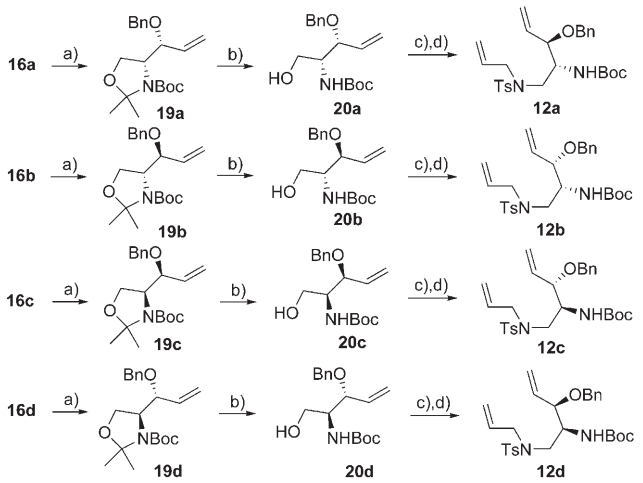
aldehyde **14** with bromo derivative **15** followed by MnO_2 oxidation and functional group interconversions, while fragments **8** and **8a** should be preparable in good yields from *p*-hydroxybenzoic acid. The hexahydroazepine and aromatic domains can be coupled by Mukaiyama esterification and amidation procedures to generate fully protected precursors for target compounds. Finally total synthesis of **1a–d**, **2a–d**, and **3** should be accomplished through global deprotection of their fully protected precursors.

Synthesis of the divinyl derivatives 11a–d and 12a–d: Our synthesis started with the preparation of diastereomerically pure alcohol precursors **16a–d** (Scheme 2). Addition of vinylmagnesium bromide to freshly prepared[64] (*R*)-Garner aldehyde (**13a**) at −78 °C delivered a mixture of *syn*-**16a** and *anti*-**16b** allylic alcohols in 1:6 diastereomeric ratio.[66] The two diastereomers were separated by flash chromatography.

Similarly, addition of vinylmagnesium bromide to (*S*)-Garner aldehyde (**13b**) furnished *syn*-**16c** and *anti*-**16d**. To enhance the yields of *syn* alcohols the major *anti* alcohols **16b** and **16d** were converted into their corresponding *syn* alcohols **16a** and **16c**, respectively, by a Mitsunobu approach.[67] Other methods to acquire *syn* alcohols as the major products by addition of vinyllithium[68] (generated in situ from tetravinyltin and methyllithium) and zinc bromide on Garner aldehyde were not suitable for scaling-up purposes, due to the lower stabilities of the reagents. Diastereomerically pure allylic alcohols **16a–d** were converted into their corresponding MOM ethers **17a–d** by treatment with MOM chloride in quantitative yields. Selective removal of the isopropylidene groups of **17a–d** with catalytic amounts of PTSA in methanol secured **18a–d**, respectively. Further tosylation of the resulting alcohols, followed by replacement of tosyloxy groups with allylamine at 65 °C in a sealed tube, furnished amines, which were protected as benzyl carbamates to afford the divinyl derivatives **11a–d** (Scheme 2).

Similarly, alcohols **16a–d** were converted into their corresponding benzyl ether derivatives **19a–d** by treatment with NaH and benzyl bromide in THF at 0 °C (Scheme 3). Opening of the isopropylidene rings of **19a–d** furnished alcohols **20a–d**, respectively, which were further tosylated. The tosyloxy derivatives of **20a–d** were treated with allylamine in sealed tube to afford amine derivatives, which were further converted into their corresponding divinyl sulfonamides **12a–d** through tosyl protection of their amino functionalities.

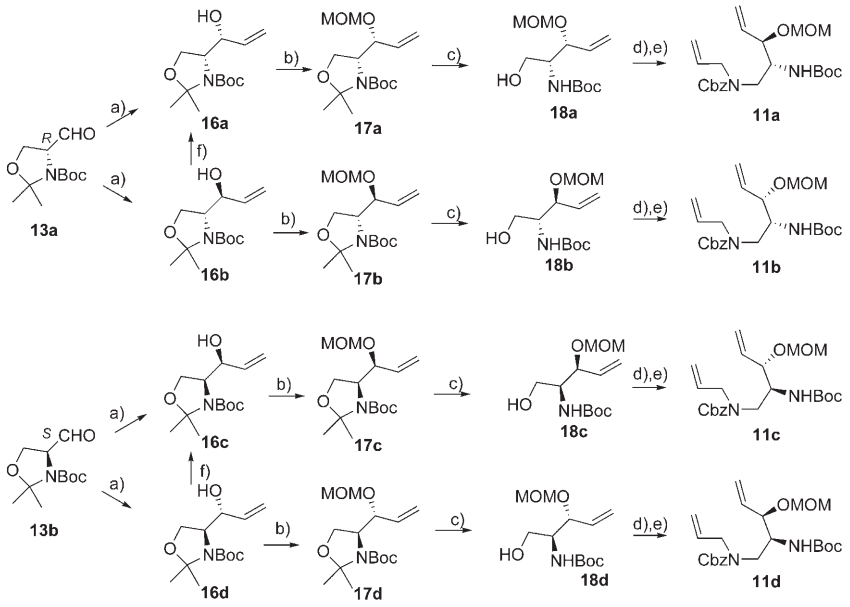
Synthesis of amido alcohols 22a–d and 24a–d: Ring-closing metatheses of the diastereomerically pure divinyl derivatives **11a–d** and **12a–d** in the presence of Grubbs' first-generation



Scheme 3. Synthesis of divinyl derivatives **12a–d**. a) BnBr, NaH, THF, 0 °C→RT; b) PTSA, MeOH, 0 °C→RT, c) i) TsCl, Et_3N, CH_2Cl_2, 0 °C, ii) allylamine, MeOH, 65 °C, d) TsCl, Et_3N, CH_2Cl_2, 0 °C.

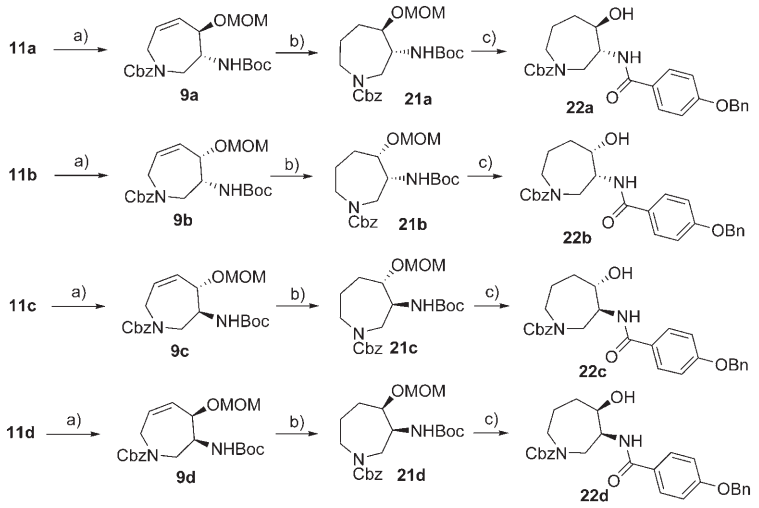
catalyst delivered the tetrahydroazepines **9a–d** and **10a–d**, respectively, in quantitative yields (Scheme 4). Reduction of the double bonds in **9a–d** by hydrogenation at atmospheric pressure in THF furnished the hexahydroazepine cores **21a–d**. Simultaneous removal of MOM and Boc with 50% TFA in CH_2Cl_2 afforded the amino alcohols, which were further coupled with *p*-benzyloxybenzoyl chloride in Et_3N/CH_2Cl_2 to provide their corresponding amido alcohols **22a–d**.

Similarly, to achieve the synthesis of amido alcohols **24a–d**, tetrahydroazepines **10a–d** were converted into **23a–d** through simultaneous Boc removal and double bond reduction followed by debenzylation with 10% Pd/C in methanol and HCl under pressure (Scheme 5). Amines **23a–d** were further coupled with *p*-benzyloxybenzoyl chloride after regeneration of amine with triethylamine in CH_2Cl_2 at 0 °C to provide amido alcohols **24a–d** in good yields.

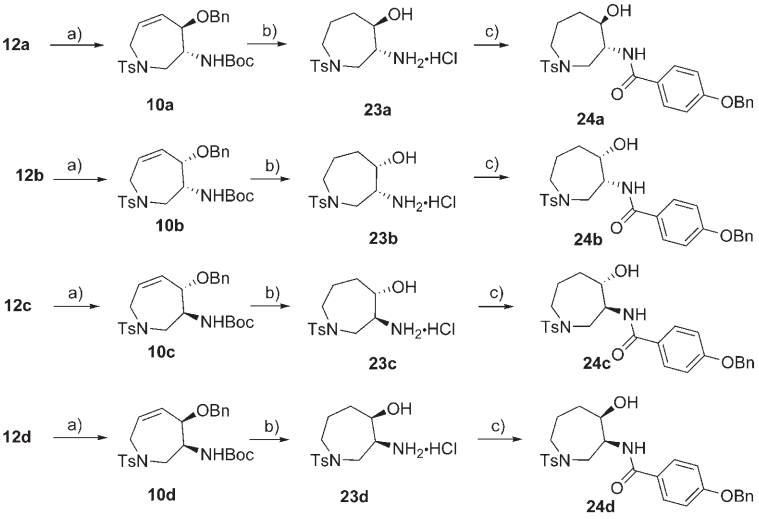


Scheme 2. Synthesis of divinyl derivatives **11a–d**. a) Vinylmagnesium bromide, THF, −78 °C; b) MOMCl, DIPEA, CH_2Cl_2, 0 °C→RT; c) PTSA, MeOH, 0 °C→RT; d) i) TsCl, Et_3N, CH_2Cl_2, 0 °C, ii) allylamine, MeOH, 65 °C; e) CbzCl, Et_3N, THF, 0 °C; f) i) *p*-nitrobenzoic acid, DEAD, PPh_3, THF; ii) K_2CO_3, MeOH, RT.

Synthesis of the benzophenone fragments 4 and 5: The benzophenone fragments of balanol and ophiocordin were synthesized by a modification of the procedure of Hollinshead et al.[69] In the reported procedure, *m*-hydroxybenzaldehyde was converted into aldehyde **29** (Scheme 6) in five steps with the use of dibromotetrafluoroethane as a brominating agent in one of the intermediate steps. To reduce the number of steps and to avoid the use of dibromotetrafluoroethane, MOM-protected alde-



Scheme 4. Synthesis of amido alcohols **22a–d**. a) Grubbs' catalyst (1st gen), CH_2Cl_2, 45 °C, b) H_2/Pd-C, THF, RT, c) i) TFA, CH_2Cl_2, RT, 4 h s, ii) *p*-benzyloxybenzoyl chloride, CH_2Cl_2, Et_3N.



Scheme 5. Synthesis of amido alcohols **24a–d**. a) Grubbs catalyst (1st gen.), CH_2Cl_2, 45 °C; b) H_2/10 % Pd-C, MeOH/HCl; c) *p*-benzyloxybenzoyl chloride, CH_2Cl_2, Et_3N, 0 °C.

hyde **14**, which can easily be synthesized in three steps from *m*-hydroxybenzaldehyde,[69] was used. Coupling of **14** with the anion generated from bromo derivative **15** provided alcohol **25**, which was oxidized to the ketone **26** with MnO_2. The ketal of **26** was unmasked with catalytic amounts of PTSA in acetone/water 9:1 to provide aldehyde **27**, which was oxidized to the acid **4** required for the synthesis of ophiocordin **3**. Acid **4** was again esterified with benzyl bromide and K_2CO_3 to furnish **28**. Selective hydrolysis of the *tert*-butyl ester component by thermolysis[38] in quinoline at 195 °C delivered acid **5**, required for the synthesis of **1a–d** and **2a–d**.

Coupling of fragments and generation of balanol and analogues: The acid **5** was further coupled with amido alcohols **22a–d** and **24a–d** by Mukaiyama esterification[70] to provide the fully protected balanol derivatives **30a–d** and **31a–d**, respectively.

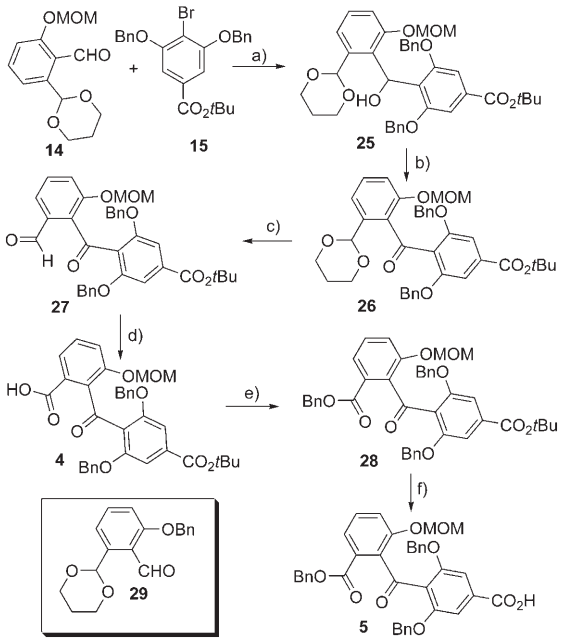
MOM removal in cat HCl/MeOH (Scheme 7), followed by debenzylation and purification of **30a–d** by Nicolaou's procedure allowed the total syntheses of (−)-balanol (**1a**) and its other stereosiomers **1b–d**. The spectral data and optical purity[46] of (−)-balanol **1a** were in accordance with the reported properties. Fully protected **31a** was converted into N-tosyl balanol **2a** (Scheme 8) by simultaneous removal of MOM and benzyl groups under conditions similar to those described above. Similarly, the syntheses of *N*-tosylbalanols **2b–d** could also be achieved from the advanced precursors **31b–d**.

Synthesis of fully protected ophiocordin (3): To achieve the synthesis of ophiocordin (**3**), hexahydroazepine **21a** was converted into **32** by selective removal of MOM with trimethylsilyl bromide, and this was further esterified with *p*-benzyloxybenzoic acid to deliver **33** by Mukaiyama esterifications (Scheme 9).[70] Boc removal in TFA/CH_2Cl_2 furnished amino ester **34**. Several initial attempts at amide coupling (DCC, DIC, HBTU, TBTU) of amine **34** with acid **4** to achieve **35** were unsuccessful, which might be due to steric hindrance.

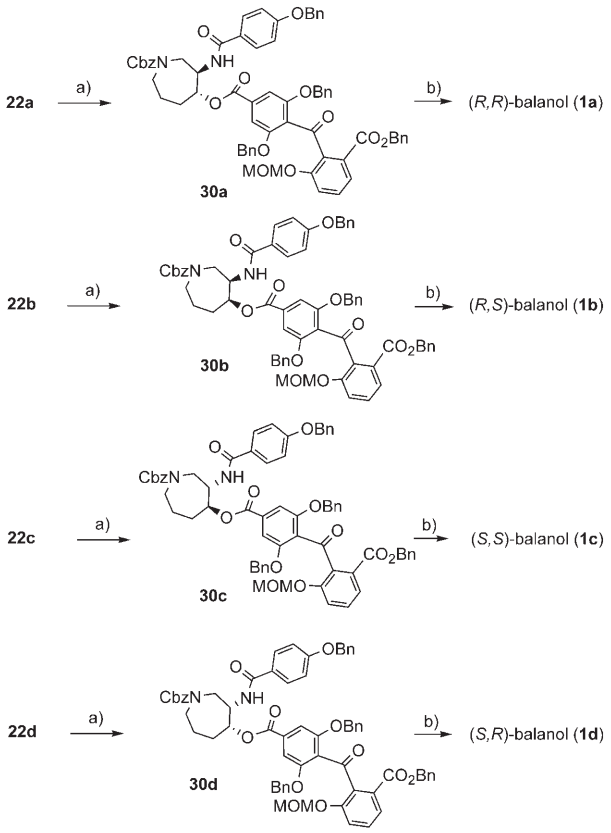
Finally, coupled product **35** was isolated by Mukaiyama amidation,[71] albeit in poor yield and with recovery of unreacted amine **34**. Ophiocordin **3** may be achieved through final deprotection of fully protected ophiocordin **35**.

Conclusion

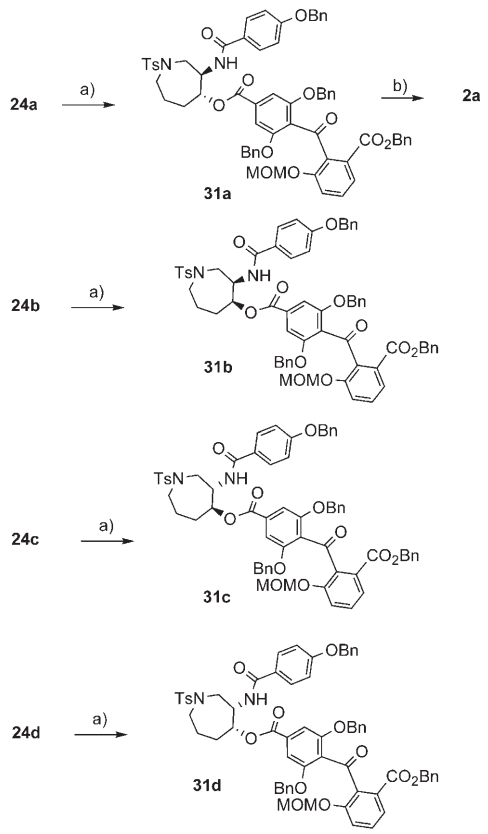
In summary, total syntheses of naturally occurring (−)-(*R*,*R*)-balanol and its stereoisomers from the easily available (*R*)- and (*S*)-Garner aldehydes have been accomplished for the first time, with overall yields of 9–13 % in a stereoselective and regioselective diversity-oriented approach. The attractiveness of the approach lies in its useful nucleophilic



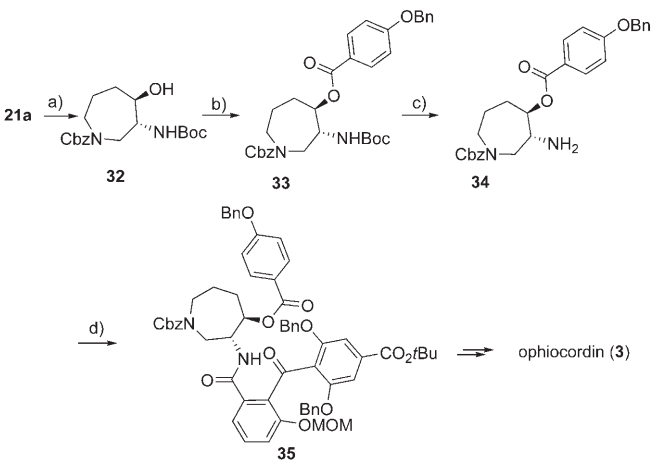
Scheme 6. Synthesis of acids **4** and **5**. a) *n*BuLi, THF, −78 °C; b) MnO_2, CH_2Cl_2; c) PTSA, acetone/water 9:1, reflux; d) NaH_2PO_4, H_2O_2, $NaClO_2$, acetonitrile, e) benzyl bromide, K_2CO_3, acetone, f) quinoline, 195 °C.



Scheme 7. Synthesis of **1a–d**. a) Acid **5**, 2-chloro-1-methylpyridinium iodide, DMAP, Et_3N, CH_2Cl_2, RT; b) i) HCl/MeOH, RT; ii) H_2, Pd/C, THF/water/acetic acid 4:1:1.



Scheme 8. Synthesis of *N*-tosyl-balanol. a) Acid **5**, 2-chloro-1-methylpyridinium iodide, DMAP, Et_3N, CH_2Cl_2, RT; b) i) HCl/MeOH, RT; ii) H_2, Pd/C, THF/water/acetic acid 4:1:1.



Scheme 9. Synthesis of fully protected ophiocordin derivative **35**. a) Trimethylsilyl bromide, CH_2Cl_2, 0 °C→RT; b) *p*-benzyloxybenzoic acid, 2-chloro-1-methylpyridinium iodide, DMAP, Et_3N, CH_2Cl_2, RT; c) TFA/CH_2Cl_2; d) acid **4**, 2-chloro-1-methylpyridinium iodide, DMAP, Et_3N, CH_2Cl_2, RT.

addition with high diastereoselectivity, the displacement reaction for access to the divinyl derivatives, and the ruthenium-catalyzed olefin metathesis for the construction of the azepine rings. This easily executed sequence gave access to the key hexahydroazepine building block units **21a–d** and

22a–d, which are well suited for the preparation of several balanol analogues for accessing more selective PKC inhibitors. The synthetic route can also be further exploited for the synthesis of other isosteres of balanols.

Experimental Section

General remarks: Melting points were determined on a COMPLAB melting point apparatus and are uncorrected. IR spectra were recorded on a Perkin–Elmer RXI FT-IR spectrometer. ^{1}H NMR and ^{13}C NMR spectra were recorded on Bruker DPX 200 (operating at 200 MHz for ^{1}H and 50 MHz for ^{13}C) or DPX 300 (operating at 300 MHz for ^{1}H and 75 MHz for ^{13}C) spectrometers in $CDCl_3$ and CD_3OD as solvents. Tetramethylsilane (0.00 ppm) served as an internal standard in ^{1}H NMR, and $CDCl_3$ (77.0 ppm) in ^{13}C NMR. All spectra were recorded at 25 °C. Coupling constants (J values) are given in Hz. Chemical shifts are expressed in parts per million (ppm). Mass spectra were recorded by electron spray ionization (ESI) or fast atom bombardment spectrometry (FAB-MS) on a JEOL SX 102 spectrometer with argon/xenon as the FAB gas. Glycerol or *m*-nitrobenzyl alcohol was used as matrix. Elemental analyses were done on Varian EL-III CHN analyzer (Germany). Reactions were monitored on silica gel TLC plates (coated with TLC-grade silica gel, obtained from Merck). Detecting agents used (for TLC) were iodine vapors and/or spraying with an aqueous solution of vanillin in 10% sulfuric acid followed by heating at 150 °C. Column chromatography was performed over silica gel (60–120 mesh) procured from Qualigens (India) with freshly distilled solvents. Anhydrous tetrahydrofuran used in Mitsunobu reactions was obtained from Spectrochem and heated at reflux over sodium/benzophenone prior to use.

(1*S*,4*R*)-4-(1-Hydroxyallyl)-2,2-dimethyloxazolidine-3-carboxylic acid *tert*-butyl ester (16b): Freshly prepared vinylmagnesium bromide (1 M solution in THF, 175 mL) was added dropwise at −78 °C to a cooled solution of (*R*)-Garner aldehyde (**13a**, 10 g, 38.9 mmol) in anhydrous THF (325 mL). The reaction mixture was stirred for 2 h at the same temperature and was then allowed to warm to room temperature before careful addition of saturated ammonium chloride solution in water. The solution was extracted with ether (500 mL × 2). The combined organic extracts were dried over anhydrous Na_2SO_4, filtered, and concentrated in vacuo. The residue was chromatographed on silica gel with ethyl acetate in hexane (15%) as eluent to furnish a mixture of **16a** and **16b** 1:6 as a colorless oil (10.6 g, 94.6%). $[\alpha]_D^{25} = +27.6$ ($c = 1.47$, chloroform). Pure **16b** was further obtained by flash chromatography on silica gel (8.5 g, 75.8%). $[\alpha]_D^{25} = +54.6$ ($c = 1.47$, chloroform); $R_f = 0.4$ (ethyl acetate in hexane, 20%); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 5.87$–5.82 (m, 1H; -C*H*=CH$_2$), 5.38 (d, $J = 18$ Hz, 1H; -CH=C*H*$_2$), 5.23 (d, $J = 12$ Hz, 1H; -CH=C*H*$_2$), 4.29–4.18 (br m, 1H; C*H*-OH), 4.07–4.02 (m, 1H; 4-*H*), 3.99–3.90 (m, 2H; 5-*H*), 1.57 (s, 3H; C*H*$_3$), 1.51 (s, 9H; OC(C*H*$_3$)$_3$}, 1.45 ppm (s, 3H; C*H*$_3$); ^{13}C NMR (50 MHz, $CDCl_3$+CCl_4, 25 °C, TMS): $\delta = 154.9$, 137.9, 117.8, 94.7, 81.1, 75.6, 65.0, 62.3, 28.7, 27.4, 24.7 ppm; IR (neat): $\tilde{\nu} = 3449$, 2982, 2363, 1696 (-NH-C=O), 1389, 1287 cm^{-1}; MS (FAB): m/z (%): 258 (60) $[M+H]^+$, 202 (100) $[M-t\text{Bu}]^+$, 184 (50) $[M-t\text{Bu}-H_2O]^+$; elemental analysis (%) calcd for $C_{13}H_{23}NO_4$: C 60.68, H 9.01, N 5.44; found: C 60.57, H 8.89, N 5.34.

(1*R*,4*R*)-4-(1-Hydroxyallyl)-2,2-dimethyloxazolidine-3-carboxylic acid *tert*-butyl ester (16a): Diethyl azodicarboxylate (5.4 g, 0.031 mol) was added dropwise at 0 °C to a stirred solution of alcohol **16b** (4.0 g, 0.015 mol), triphenylphosphine (8.34 g, 0.031 mol), and 4-nitrobenzoic acid (5.2 g, 0.031 mol) in anhydrous THF (100 mL). The reaction mixture was stirred at 0 °C for 15 min and then for 1 h at room temperature. The reaction mixture was concentrated and chromatographed over silica gel to afford the nitrobenzoate as a syrup. Solid K_2CO_3 (3.1 g, 0.022 mol) was added to a solution of the nitrobenzoate (4.5 g, 0.011 mol) in methanol (50 mL) and the reaction mixture was stirred for 30 min at room temperature. After completion of the reaction, solid was filtered off, and the filtrate was concentrated in vacuo. The residue was diluted with ethyl acetate, washed with water followed by brine, dried over anhydrous Na_2SO_4, and concentrated in vacuo. The residue was chromatographed over silica gel with ethyl acetate in hexane (15%) as eluent to furnish **16a** (2.70 g, 92%) as a white solid. $R_f = 0.4$ (ethyl acetate in hexane, 20%); $[\alpha]_D^{25} = +47.9$ ($c = 1.25$ in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 5.89$–5.82 (m, 1H; -C*H*=CH$_2$), 5.85 (d, $J = 18$ Hz, 1H; -CH=C*H*$_2$), 5.30 (d, $J = 12$ Hz, 1H; -CH=C*H*$_2$), 4.28 (pseudo t, 1H; C*H*-OH), 4.19 (br m, 1H; 4-*H*), 4.07–3.90 (m, 2H; 5-*H*), 1.60 (s, 3H; C*H*$_3$), 1.57 (s, 9H; OC(C*H*$_3$)$_3$), 1.46 ppm (s, 3H; C*H*$_3$); ^{13}C NMR (75 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 154.9$ (C=O), 137.9 (-*C*H=CH$_2$), 117.9 (CH=*C*H$_2$), 96.5 (C-2), 81.2 (-*C*H-OH), 74.3 (*C*(CH$_3$)$_3$), 65.0 (C-5), 62.3 (C-4), 28.8 (*C*(CH$_3$)$_3$), 26.7 ppm (*C*H$_3$); IR (neat): $\tilde{\nu} = 3450$, 2981, 2363, 1697 (-NH-C=O), 1382, 1260 cm^{-1}; MS (FAB): m/z (%): 258 (80) $[M+H]^+$, 202 (100) $[M-t\text{Bu}]^+$, 184 (65) $[M-t\text{Bu}-H_2O]^+$; elemental analysis (%) calcd for $C_{13}H_{23}NO_4$: C 60.68, H 9.01, N 5.44; found: C 60.64, H 8.81, N 5.29.

(1*R*,4*S*)-4-(1-Hydroxyallyl)-2,2-dimethyloxazolidine-3-carboxylic acid *tert*-butyl ester (16d): This compound was prepared as described for **16b** from (*S*)-Garner aldehyde (**13b**). Yield = 74.3%; $R_f = 0.4$ (ethyl acetate in hexane, 20%); $[\alpha]_D^{25} = -32.3$ ($c = 2.1$ in methanol); ^{1}H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 5.91$–5.68 (m, 1H; -C*H*=CH$_2$), 5.41–5.13 (m, 2H; -CH=C*H*$_2$), 4.39 (br s, 1H; C*H*-OH), 4.22 (br s, 1H; 4-*H*), 4.16–3.81 (m, 2H; 5-*H*), 1.55 (s, 3H; C*H*$_3$), 1.50 (s, 9H; OC(C*H*$_3$)$_3$), 1.48 ppm (s, 3H; C*H*$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 155.4$, 137.1, 116.6, 94.9, 81.5, 74.6, 65.0, 62.4, 28.7, 26.6, 24.8 ppm; IR (neat): $\tilde{\nu} = 3456$, 2981, 2345, 1689 (-NH-C=O), 1376, 1280 cm^{-1}; MS (ESI): m/z (%): 257 (100) $[M]^+$, 202 (50) $[M-t\text{Bu}]^+$, 184 (25) $[M-t\text{Bu}-H_2O]^+$; elemental analysis (%) calcd for $C_{13}H_{23}NO_4$: C 60.68, H 9.01, N 5.44; found: C 60.49, H 8.82, N 5.45.

(1*S*,4*S*)-4-(1-Hydroxyallyl)-2,2-dimethyloxazolidine-3-carboxylic acid *tert*-butyl ester (16c): This compound was prepared as described for **16a** from **16d**. Yield = 63%; $R_f = 0.4$ (ethyl acetate in hexane, 20%); $[\alpha]_D^{25} = -54.3$ ($c = 2.1$ in methanol); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 5.88$–5.81 (m, 1H; -C*H*=CH$_2$), 5.41–5.31 (m, 1H; -CH=C*H*$_2$), 5.22 (d, $J = 10.2$ Hz, 1H; -CH=C*H*$_2$), 4.29 (br s, 1H; C*H*-OH), 4.18 (s, 1H; 4-*H*), 4.07–3.56 (m, 2H; 5-*H*), 1.56 (s, 3H; C*H*$_3$), 1.51 (s, 9H; OC(C*H*$_3$)$_3$), 1.46 ppm (s, 3H; C*H*$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 155.3$, 137.9, 118.1, 94.8, 81.4, 76.0, 65.6, 62.1, 59.7, 28.7, 27.4, 26.7 ppm; IR (neat): $\tilde{\nu} = 3467$, 2981, 2363, 1691 (-NH-C=O), 1379, 1280 cm^{-1}; MS (FAB): m/z (%): 280 (100) $[M+Na]^+$, 202 (35) $[M-t\text{Bu}]^+$, 184 (50) $[M-t\text{Bu}-H_2O]^+$; elemental analysis (%) calcd for $C_{13}H_{23}NO_4$: C 60.68, H 9.01, N 5.44; found: C 60.51, H 8.89, N 5.26.

General Procedure for MOM protection: DIPEA (1.35 mL, 7.78 mmol) was added at 0 °C to a cooled solution of the alcohol (1.0 g, 3.89 mmol) in dry CH_2Cl_2 (15 mL), followed by MOM chloride (0.44 mL, 5.84 mmol). The reaction mixture was stirred at the same temperature for 4 h. After completion of the reaction, solvent was evaporated, and the residue was diluted with ethyl acetate. The organic layer was washed with water and brine, dried over anhydrous Na_2SO_4, and concentrated in vacuo. The residue was chromatographed over silica gel with ethyl acetate in hexane (5%) as eluent to furnish the protected alcohol.

(1*R*,4*R*)-4-(1-Methoxymethoxyallyl)-2,2-dimethyloxazolidine-3-carboxylic acid *tert*-butyl ester (17a): Yield = 84%; $R_f = 0.62$ (ethyl acetate in hexane, 10%); $[\alpha]_D^{25} = +72.4$ ($c = 0.915$ in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 5.77$–5.71 (m, 1H; -C*H*=CH$_2$), 5.31–5.22 (m, 2H; -CH=C*H*$_2$), 4.71 (d, $J = 6.6$ Hz, 1H; -C*H*$_2$-O-CH$_3$), 4.56 (d, $J = 6.6$ Hz, 1H; -C*H*$_2$-O-CH$_3$), 4.39–4.28 (br m, 1H; 4-*H*), 4.08 (dd, $J = 8.4$,1.2 Hz, 1H; -C*H*-OMOM), 4.01–3.87 (m, 2H; 5-*H*), 3.35 (s, 3H; OC*H*$_3$), 1.51 (s, 3H; C*H*$_3$), 1.49 (s, 3H; C*H*$_3$), 1.48 ppm (s, 9H; OC(C*H*$_3$)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 151.2$ (C=O), 134.3 (-*C*H=CH$_2$), 117.9 (-CH=*C*H$_2$), 93 (O-*C*H$_2$-O), 78.7 (*C*H-OMOM), 62.7 (*C*(CH$_3$)$_3$), 58.9 (C-4), 57.9 (O-*C*H$_3$), 54.6 (C-5), 28.4 (*C*(CH$_3$)$_3$), 27.1 (*C*H$_3$), 25.6 ppm (*C*H$_3$); IR (neat): $\tilde{\nu} = 3861$, 3821, 2932, 2362, 1700, 1386, 1169, 1098, 1033 cm^{-1}; MS (ESI): m/z (%): 301.9 (89) $[M]^+$, 245.9 (100) $[M-t\text{Bu}]^+$, 202.1 (12) $[M-t\text{Boc}]^+$; elemental analysis (%) calcd for $C_{15}H_{27}NO_5$: C 59.78, H 9.03, N 4.65; found: C 59.68, H 8.93, N 4.56.

(1*S*,4*R*)-4-(1-Methoxymethoxyallyl)-2,2-dimethyloxazolidine-3-carboxylic acid *tert*-butyl ester (17b): Yield = 81%; $R_f = 0.64$ (ethyl acetate in hexane, 10%); $[\alpha]_D^{25} = +61.4$ ($c = 0.376$ in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 5.76$–5.68 (m, 1H; -C*H*=CH$_2$), 5.35–5.22 (m, 2H;

-CH=CH_2), 4.63 (d, J=6.6 Hz, 1H; -CH_2-O-CH$_3$), 4.56 (d, J=6.6 Hz, 1H; -CH_2-O-CH$_3$), 4.41–4.26 (br m, 1H; -CH-OMOM), 4.09–3.76 (m, 3H; 4-H and 5-H), 3.37 (s, 3H; OCH_3), 1.61 (s, 3H; CH_3), 1.51 (s, 3H; CH_3), 1.45 ppm (s, 9H; OC(CH_3)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): δ=152.7, 135.9, 120.5, 94.1, 85.5, 80.2, 70.2, 64.7, 60.2, 55.9, 28.6, 27.7, 26.6 ppm; IR (neat): $\tilde{\nu}$=3860, 3821, 2912, 2361, 1710, 1386, 1169, 1068, 1033 cm^{-1}; MS (ESI): m/z (%): 301.9 (89) $[M]^+$, 245.9 (100) $[M-t\text{Bu}]^+$, 202.1 (12) $[M-t\text{Boc}]^+$; elemental analysis (%) calcd for $C_{15}H_{27}NO_5$: C 59.78, H 9.03, N 4.65; found: C 59.71, H 8.90, N 4.51.

(1*S*,4*S*)-4-(1-Methoxymethoxyallyl)-2,2-dimethyloxazolidine-3-carboxylic acid *tert*-butyl ester (17c): Yield=82%; R_f=0.6 (ethyl acetate in hexane, 10%) $[\alpha]_D^{25}$=−77.4 (c=0.715 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ=5.74–5.67 (m, 1H; -CH=CH$_2$), 5.31–5.22 (m, 2H; -CH=CH_2), 4.71 (d, J=6.6 Hz, 1H; -CH_2-O-CH$_3$), 4.56 (d, J=6.6 Hz, 1H; -CH_2-O-CH$_3$), 4.39–4.28 (br m, 1H; 4-H), 4.08 (dd, J=8.4, 1.2 Hz, 1H; -CH-OMOM), 4.01–3.87 (m, 2H; 5-H), 3.35 (s, 3H; O-CH_3), 1.51 (s, 3H; CH_3), 1.49 (s, 3H; CH_3), 1.46 ppm (s, 9H; OC(CH_3)$_3$); ^{13}C NMR (75 MHz, $CDCl_3$, 25 °C, TMS): δ=151.1, 134.3, 127.4, 117.8, 93.0, 88.1, 78.6, 72.1, 63.1, 58.6, 54.4, 27.0, 27.5 ppm; IR (neat): $\tilde{\nu}$=3851, 3821, 2942, 2262, 1732, 1380, 1098, 1033 cm^{-1}; MS (ESI): m/z (%): 301.9 (89) $[M]^+$, 245.9 (100) $[M-t\text{Bu}]^+$, 202.1 (12) $[M-t\text{Boc}]^+$; elemental analysis (%) calcd for $C_{15}H_{27}NO_5$: C 59.78, H 9.03, N 4.65; found: C 59.69, H 8.89, N 4.60.

(1*R*,4*S*)-4-(1-Methoxymethoxy-allyl)-2,2-dimethyl-oxazolidine-3-carboxylic acid *tert*-butyl ester (17d): Yield=83.4%; R_f=0.6 (ethyl acetate in hexane, 10%); $[\alpha]_D^{25}$=−66.4 (c=0.776 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ=5.75–5.70 (m, 1H), 5.30–5.24 (m, 2H), 4.68 (d, J=6.6 Hz, 1H), 4.53 (d, J=6.6 Hz, 1H), 4.38–4.35 (br m, 1H), 4.13–3.88 (m, 3H), 3.34 (s, 3H; OCH_3), 1.55 (s, 3H; -CH_3), 1.48 (s, 3H; -CH_3), 1.44 ppm (s, 9H; OC(CH_3)$_3$); ^{13}C NMR (75 MHz, $CDCl_3$, 25 °C, TMS): δ=151.1, 134.3, 117.9, 92.6, 78.8, 64.9, 62.7, 58.8, 54.4, 50.9, 27.0, 25.6 ppm; IR (neat): $\tilde{\nu}$=3900, 3811, 2932, 2342, 1700, 1356, 1160, 1098, 1033 cm^{-1}; MS (ESI): m/z (%): 301.6 (79) $[M]^+$, 245.6 (100) $[M-t\text{Bu}]^+$, 202.1 (32) $[M-t\text{Boc}]^+$; elemental analysis (%) calcd for $C_{15}H_{27}NO_5$: C 59.78, H 9.03, N 4.65; found: C 59.73, H 8.93, N 4.59.

General Procedure for benzyl protection: Washed NaH (1.86 g, 7.78 mmol) was added at 0 °C to a cooled solution of the alcohol (1.0 g, 3.89 mmol) in dry THF (15 mL), followed by benzyl bromide (800 mg, 4.67 mmol). The reaction mixture was stirred at room temperature for 5 h. After completion of the reaction, methanol was added dropwise to neutralize excess NaH, and solvent was evaporated. The residue was diluted with ethyl acetate, washed with water followed by brine, dried over anhydrous Na_2SO_4, and concentrated in vacuo. The residue was chromatographed over silica gel to furnish the protected product.

(4*R*,6*R*)-4-(1-Benzyloxyallyl)-2,2-dimethyloxazolidine-3-carboxylic acid *tert*-butyl ester (19a): Yield=78%; R_f=0.5 (ethyl acetate in hexane, 6%); $[\alpha]_D^{25}$=+59.4 (c=0.711 in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): δ=7.32 (s, 5H; ArH), 5.96–5.61 (m, 1H; -CH=CH$_2$), 5.32–5.24 (m, 2H; -CH=CH_2), 4.62 (d, J=11.6 Hz, 1H; -CH_2-Ph), 4.36 (d, J=11.6 Hz, 1H; -CH_2-Ph), 4.11–4.03 (m, 2H; 4-H, -CHOBn), 3.90 (br s, 2H; 5-H), 1.59 (s, 3H; -CH_3), 1.51 (s, 3H; -CH_3), 1.46 ppm (s, 9H; -OC-(CH_3)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$): δ=152.8 (C=O), 138.3 (-CH=CH$_2$), 136.4, 128.1, 128.3, 119.7, 118.1 (-CH=CH_2), 94.3 (C-2), 81.4, 80.4, 71.2 (C(CH$_3$)$_3$), 65.6, 64.8, 60.2, 60.8 (C-4), 28.1, 27.4, 25.9, 23.1 ppm; IR (neat): $\tilde{\nu}$=3984, 3876, 3712, 3537, 2979, 1838, 1699, 1384, 1170, 1084 cm^{-1}; MS (FAB): m/z (%): 348 (29) $[M+\text{H}]^+$, 292 (45) $[M-t\text{Bu}]^+$, 248 (51) $[M-t\text{Boc}]^+$, 234 (85) $[M-\text{Bn}+\text{Na}]^+$, 91 (100) $[\text{PhCH}_2]^+$; elemental analysis (%) calcd for $C_{20}H_{29}NO_4$: C 69.14, H 8.41, N 4.03; found: C 68.97, H 8.34, N 3.99.

(4*R*,6*S*)-4-(1-Benzyloxyallyl)-2,2-dimethyloxazolidine-3-carboxylic acid *tert*-butyl ester (19b): Yield=76.8%; R_f=0.51 (ethyl acetate in hexane, 6%); $[\alpha]_D^{25}$=+46.4 (c=0.738 in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): δ=7.27 (s, 5H; ArH), 5.91–5.67 (m, 1H; -CH=CH$_2$), 5.30–5.21 (m, 2H; -CH=CH_2), 4.59 (d, J=11.6 Hz, 1H; -CH_2-Ph), 4.33 (d, J=11.6 Hz, 1H; -CH_2-Ph), 4.11–3.89 (m, 4H; 4-H, 5-H, -CHOBn), 1.51 (s, 3H; -(CH_3)$_2$), 1.46 (s, 9H; -(CH_3)$_2$), 1.43 ppm (s, 3H; -OC(CH_3)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$+CCl_4): δ=152.7 (C=O), 138.7 (-CH=CH$_2$), 136.9, 135.5, 128.7, 120.6, 119.4 (-CH=CH_2), 94.1, 81.6, 80.0, 79.4, 71.2, 65.5, 64.0, 60.6, 28.8, 27.3, 25.4 ppm; IR (neat): $\tilde{\nu}$=3984, 3876, 3712, 3537, 2979, 1838, 1699, 1384, 1170, 1084 cm^{-1}; MS (FAB): m/z (%): 348 (27) $[M+\text{H}]^+$, 292 (65) $[M-t\text{Bu}]^+$, 248 (100) $[M-t\text{Boc}]^+$; elemental analysis (%) calcd for $C_{20}H_{29}NO_4$: C 69.14, H 8.41, N 4.03; found: C 68.87, H 8.04, N 3.96.

(4*S*,6*S*)-4-(1-Benzyloxyallyl)-2,2-dimethyloxazolidine-3-carboxylic acid *tert*-butyl ester (19c): Yield=79%; R_f=0.6 (ethyl acetate in hexane, 10%); $[\alpha]_D^{25}$=−53.4 (c=1.714 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ=7.34 (s, 5H; ArH), 5.89–5.78 (m, 1H; -CH=CH$_2$), 5.33–5.26 (m, 2H; -CH=CH_2), 4.63 (d, J=11.6 Hz, 1H; -CH_2-Ph), 4.33 (d, J=11.6 Hz, 1H; -CH_2-Ph), 4.16–3.98 (m, 2H; 4-H, -CHOBn), 3.92–3.88 (m, 2H; 5-H), 1.51 (s, 3H; -CH_3), 1.46 (s, 9H; -OC(CH_3)$_3$), 1.43 ppm (s, 3H; -CH_3); ^{13}C NMR (50 MHz, $CDCl_3$): δ=152.9 (C=O), 138.8 (-CH=CH$_2$), 136.8, 128.8, 128.3, 119.7, 118.9 (-CH=CH_2), 94.6 (C-2), 81.7, 80.6, 71.2 (C(CH$_3$)$_3$), 65.6, 64.8, 60.6, 60.4 (C-4), 28.8, 27.4, 25.4, 23.6 ppm; IR (neat): $\tilde{\nu}$=3981, 3876, 3612, 3537, 2979, 1830, 1702, 1384, 1170, 1084 cm^{-1}; MS (ESI): m/z (%): 360.3 (29) $[M+\text{Na}]^+$, 292.3 (45) $[M-t\text{Bu}]^+$, 248.3 (100) $[M-t\text{Boc}]^+$; elemental analysis (%) calcd for $C_{20}H_{29}NO_4$: C 69.14, H 8.41, N 4.03; found: C 69.00, H 8.24, N 3.79.

(4*S*,6*R*)-4-(1-Benzyloxyallyl)-2,2-dimethyloxazolidine-3-carboxylic acid *tert*-butyl ester (19d): Yield=78.5%; R_f=0.54 (ethyl acetate in hexane, 8%); $[\alpha]_D^{25}$=−49.8 (c=2.025 in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): δ=7.32 (s, 5H; ArH), 5.89–5.78 (m, 2H; -CH=CH$_2$), 5.32–5.23 (m, 1H; -CH=CH_2), 4.61 (d, J=11.6 Hz, 1H; -CH_2-Ph), 4.35 (d, J=11.6 Hz, 1H; -CH_2-Ph), 4.15–4.02 (m, 2H; 4-H, -CHOBn), 3.90 (br s, 2H; 5-H), 1.59 (s, 3H; -CH_3), 1.49 (s, 3H; -CH_3), 1.46 ppm (s, 9H; -OC-(CH_3)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): δ=152.9, 138.8, 136.8, 128.7, 128.3, 119.6, 118.8, 94.6, 81.6, 80.4, 71.2, 65.6, 64.7, 60.6, 30.1, 28.8, 27.3 ppm; IR (neat): $\tilde{\nu}$=3967, 3871, 3712, 3537, 2979, 1838, 1699, 1374, 1170, 1084 cm^{-1}; MS (FAB): m/z (%): 348 (29) $[M+\text{H}]^+$, 292 (45) $[M-t\text{Bu}]^+$, 248 (51) $[M-t\text{Boc}]^+$, 234 (85) $[M-\text{Bn}+\text{Na}]^+$, 91 (100) $[\text{PhCH}_2]^+$; elemental analysis (%) calcd for $C_{20}H_{29}NO_4$: C 69.14, H 8.41, N 4.03; found: C 68.94, H 8.24, N 3.94.

General Procedure for acetonide deprotection: A catalytic amount of PTSA (100 mg) was added to a solution of an acetonide (0.9 g, 2.99 mmol) in methanol (10 mL) and the reaction mixture was stirred at room temperature for 3 h. Methanol was removed at low temperature, the residue was dissolved in ether, and the organic layer was washed with $NaHCO_3$ (5%) followed by brine, dried over Na_2SO_4, and concentrated in vacuo. The residue was chromatographed on silica gel with ethyl acetate in hexane (40%) as eluent to furnish the deprotected product.

(1*R*,2*R*)-(1-Hydroxymethyl-2-methoxymethoxybut-3-enyl)-carbamic acid *tert*-butyl ester (18a): Yield=87%; R_f=0.33 (ethyl acetate in hexane, 30%); $[\alpha]_D^{25}$=+82.8 (c=0.433 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ=5.83–5.71 (m, 1H; -CH=CH$_2$), 5.37–5.23 (m, 3H; -CH=CH_2, NH), 4.61 (dd, J=23.7, 6.6 Hz, 2H; OCH_2OCH$_3$), 4.28 (pseudo t, J=5.4 Hz, 1H; CH-OMOM), 3.91 (dd, J=11.7, 4.2 Hz, 1H; 1-H), 3.64 (d, J=9.6 Hz, 2H; CH_2OH), 3.39 (s, 3H; OCH_3), 2.79 (br s, 1H; OH), 1.45 ppm (s, 9H; OC(CH_3)$_3$); ^{13}C NMR (75 MHz, $CDCl_3$, 25 °C, TMS): δ=154.5 (C=O), 133.6, 117.7, 94.8, 93.3, 78.2, 77.5, 60.7, 54.4, 53.4, 27.1 ppm; IR (neat): $\tilde{\nu}$=3446, 2977, 2363, 1634 cm^{-1}; MS (FAB): m/z (%): 261 (43) $[M+\text{H}]^+$, 261 (100) $[M]^+$, 205 (80) $[M-t\text{Bu}]^+$, 162 (65) $[M-t\text{Boc}]^+$; elemental analysis (%) calcd for $C_{12}H_{23}NO_5$: C 55.16, H 8.87, N 5.36; found: C 55.10, H 8.78, N 5.29.

(1*R*,2*S*)-(1-Hydroxymethyl-2-methoxymethoxy-but-3-enyl)-carbamic acid *tert*-butyl ester (18b): Yield=84%; R_f=0.31 (ethyl acetate in hexane, 30%), $[\alpha]_D^{25}$=+89.8 (c=0.476 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ=5.83–5.72 (m, 1H; -CH=CH$_2$), 5.37–5.28 (m, 3H; -CH=CH_2, NH), 4.61 (dd, J=23.7, 6.6 Hz, 2H; OCH_2OCH$_3$), 4.30 (pseudo t, J=5.4 Hz, 1H; CH-OMOM), 3.91 (dd, J=11.7, 4.2 Hz, 1H; 1-H), 3.64 (d, J=9.6 Hz, 2H; CH_2OH), 3.39 (s, 3H; OCH_3), 2.80 (br s, 1H; OH), 1.45 ppm (s, 9H; OC(CH_3)$_3$); ^{13}C NMR (75 MHz, $CDCl_3$, 25 °C, TMS): δ=154.8, 133.2, 117.7, 93.4, 78.3, 77.7, 60.8, 54.5, 53.4, 28.3, 27.0 ppm; MS (ESI): m/z (%): 284.1 (48) $[M+\text{Na}]^+$, 261.9 (100) $[M]^+$, 205.9 (86) $[M-t\text{Bu}]^+$, 162.0 (68) $[M-t\text{Boc}]^+$; elemental analysis (%) calcd for $C_{12}H_{23}NO_5$: C 55.16, H 8.87, N 5.36; found: C 55.10, H 8.78, N 5.29.

(1*S*,2*S*)-(1-Hydroxymethyl-2-methoxymethoxy-but-3-enyl)-carbamic acid *tert*-butyl ester (18c): Yield=86%; R_f=0.33 (ethyl acetate in hexane,

30%); $[\alpha]_D^{25}=-59.7$ ($c=0.438$ in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25°C, TMS): $\delta=5.87$–5.69 (m, 1H; -C*H*=CH_2), 5.39–5.29 (m, 3H; -CH=CH_2, N*H*), 4.61 (dd, $J=23.7$, 6.6 Hz, 2H; OCH_2OCH$_3$), 4.29 (pseudo t, $J=5.4$ Hz, 1H; C*H*-OMOM), 3.93 (d, $J=8.88$ Hz, 1H; 1-*H*), 3.68 (d, $J=7.42$ Hz, 2H; CH_2OH), 3.40 (s, 3H; OCH_3), 2.90 (brs, 1H; O*H*), 1.45 ppm (s, 9H; OC(CH_3)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25°C, TMS): $\delta=155.8$, 134.7, 119.0, 94.5, 79.6, 78.7, 61.9, 55.7, 54.6, 28.3 ppm; MS (ESI): *m*/*z* (%): 284.1 (43) $[M+Na]^+$, 261.9 (100) $[M]^+$, 205.9 (82) $[M-t\text{Bu}]^+$, 162.0 (79) $[M-t\text{Boc}]^+$; elemental analysis (%) calcd for $C_{12}H_{23}NO_5$: C 55.16, H 8.87, N 5.36; found: C 55.10, H 8.78, N 5.29.

(1*S*,2*R*)-(1-Hydroxymethyl-2-methoxymethoxy-but-3-enyl)-carbamic acid *tert*-butyl ester (18d): Yield = 89%; $R_f=0.31$ (ethyl acetate in hexane, 30%); $[\alpha]_D^{25}=-65.4$ ($c=0.384$ in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): $\delta=5.84$–5.73 (m, 1H; -C*H*=CH_2), 5.38–5.30 (m, 3H; -CH=CH_2, N*H*), 4.61 (dd, $J=23.7$, 6.6 Hz, 2H; OCH_2OCH$_3$), 4.28 (pseudo t, $J=5.4$ Hz, 1H; C*H*-OMOM), 3.94 (d, $J=10.7$ Hz, 1H; 1-*H*), 3.79–3.64 (m, 2H; CH_2OH), 3.39 (s, 3H; OCH_3), 2.76 (brs, 1H; O*H*), 1.45 ppm (s, 9H; OC(CH_3)$_3$); ^{13}C NMR (75 MHz, $CDCl_3$, 25°C, TMS): $\delta=154.6$, 133.4, 117.8, 93.3, 78.3, 77.6, 60.8, 54.5, 53.3, 27.1 ppm; MS (ESI): *m*/*z* (%): 284.1 (43) $[M+Na]^+$, 261.9 (100) $[M]^+$, 205.9 (82) $[M-t\text{Bu}]^+$, 162.0 (79) $[M-t\text{Boc}]^+$; elemental analysis (%) calcd for $C_{12}H_{23}NO_5$: C 55.16, H 8.87, N 5.36; found: C 55.10, H 8.78, N 5.29.

(1*R*,2*R*)-(2-Benzyloxy-1-hydroxymethyl-but-3-enyl)-carbamic acid *tert*-butyl ester (20a): Yield = 81%; $R_f=0.4$ (ethyl acetate in hexane, 40%); $[\alpha]_D^{25}=+44.6$ ($c=0.967$ in $CHCl_3$); ^{1}H NMR ($CDCl_3$, 200 MHz, 25°C, TMS): $\delta=7.32$ (s, 5H; Ar*H*), 5.92–5.74 (m, 1H; -C*H*=CH_2), 5.41–5.34 (m, 2H; -CH=CH_2), 5.27 (s, 1H; N*H*), 4.64 (d, 1H; $J=11.8$ Hz, -CH_2Ph), 4.33 (d, $J=11.8$ Hz, 1H; -CH_2Ph), 4.07 (brs, 1H; 1-*H*), 3.94 (d, $J=10.05$ Hz, 1H; 2-*H*), 3.67–3.61 (m, 2H; CH_2OH), 1.42 ppm (s, 9H; -OC(CH_3)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25°C, TMS): $\delta=155.8$, 137.6, 135.1, 128.5, 127.8, 119.2, 82.0, 79.5, 71.1, 61.9, 54.5, 28.3 ppm; IR (neat): $\tilde{\nu}=$ 3963, 3857, 3697, 1813, 1706, 1512, 1371, 1170, 1056, 771 cm^{-1}; MS (FAB): *m*/*z* (%): 330 (52) $[M+Na]^+$, 252 (98) $[M-t\text{Bu}]^+$, 208 (82) $[M-t\text{Boc}]^+$, 91 (100) $[PhCH_2]^+$; elemental analysis (%) calcd for $C_{17}H_{25}NO_4$: C 66.43, H 8.20, N 4.56; found: C 66.32, H 8.04, N 4.36.

(1*R*,2*S*)-(2-Benzyloxy-1-hydroxymethyl-but-3-enyl)-carbamic acid *tert*-butyl ester (20b): Yield = 86.5%; $R_f=0.4$ (ethyl acetate in hexane, 40%); $[\alpha]_D^{25}=+54.8$ ($c=0.967$ in $CHCl_3$); ^{1}H NMR ($CDCl_3$, 200 MHz, 25°C, TMS): $\delta=7.73$–7.25 (m, 5H; Ar*H*), 5.90–5.72 (m, 1H; -C*H*=CH_2), 5.45–5.18 (m, 2H; -CH=CH_2), 4.60 (d, $J=11.6$ Hz, 1H; -CH_2-Ph), 4.33 (d, $J=11.6$ Hz, 1H; -CH_2-Ph), 4.10 (brs, 1H), 3.97 (d, $J=10$ Hz, 1H; 2-H),3.61 (brs, 2H; -CH_2OH), 1.42 ppm (s, 9H; -OC(CH_3)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$+CCl_4, 25°C, TMS): $\delta=156.03$ (C=O), 138.11, 135.75, 128.88, 128.21, 128.13, 127.27, 119.47, 82.29, 79.73, 71.53, 71.01, 62.22, 55.02, 28.81 cm^{-1}; MS (FAB): *m*/*z* (%): 308 (52) $[M+H]^+$, 252 (85) $[M-t\text{Bu}]^+$, 208 (60) $[M-t\text{Boc}]^+$, 91 (100) $[PhCH_2]^+$; elemental analysis (%) calcd for $C_{17}H_{25}NO_4$: C 66.43, H 8.20, N 4.56; found: C 66.39, H 8.11, N 4.31.

(1*S*,2*S*)-(2-Benzyloxy-1-hydroxymethyl-but-3-enyl)-carbamic acid *tert*-butyl ester (20c): Yield = 84%; $R_f=0.4$ (ethyl acetate in hexane, 40%); $[\alpha]_D^{25}=-49.3$ ($c=0.967$ in $CHCl_3$); ^{1}H NMR ($CDCl_3$, 200 MHz, 25°C, TMS): $\delta=7.37$–7.26 (m, 5H; Ar*H*), 5.89–5.77 (m, 1H), 5.41–5.27 (m, 3H), 4.64 (d, $J=11.2$ Hz, 2H), 4.33 (d, $J=11.2$ Hz, 1H), 4.07 (s, 1H), 3.94 (d, $J=8.6$ Hz, 1H), 3.67–3.62 (m, 2H), 1.42 ppm (s, 9H; -OC(CH_3)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25°C, TMS): $\delta=155.8$, 137.6, 135.0, 128.5, 127.8, 119.2, 82.0, 79.5, 71.0, 61.9, 54.4, 28.3 ppm; MS (FAB): *m*/*z* (%): 330 (52) $[M+Na]^+$, 252 (98) $[M-t\text{Bu}]^+$, 208 (82) $[M-t\text{Boc}]^+$, 91 (100) $[PhCH_2]^+$; elemental analysis (%) calcd for $C_{17}H_{25}NO_4$: C 66.43, H 8.20, N 4.56; found: C 66.22, H 8.14, N 4.39.

(1*S*,2*R*)-(2-Benzyloxy-1-hydroxymethyl-but-3-enyl)-carbamic acid *tert*-butyl ester (20d): Yield = 88%; $R_f=0.4$ (ethyl acetate in hexane, 40%); $[\alpha]_D^{25}=-51.8$ ($c=0.953$ in $CHCl_3$); ^{1}H NMR ($CDCl_3$, 300 MHz, 25°C, TMS): $\delta=7.37$–7.33 (m, 5H), 5.91–5.79 (m, 1H), 5.43–5.32 (m, 2H), 4.67 (d, $J=11.2$ Hz, 1H), 4.35 (d, $J=11.2$ Hz, 1H), 4.12 (s, 1H), 4.00–3.96 (brm, 1H), 3.73–3.64 (m, 2H), 1.45 ppm (s, 9H; -OC(CH_3)$_3$); ^{13}C NMR (75 MHz, $CDCl_3$): $\delta=154.5$ (C=O), 133.6, 135.0, 128.5, 127.9, 119.2, 82.2,79.1, 71.2, 62.1, 54.4, 28.3 ppm; MS (ESI): *m*/*z* (%): 330.1 (52) $[M+Na]^+$, 252.1 (80) $[M-t\text{Bu}]^+$, 208 (100) $[M-t\text{Boc}]^+$; elemental analysis (%) calcd for $C_{17}H_{25}NO_4$: C 66.43, H 8.20, N 4.56; found: C 66.34, H 8.12, N 4.44.

General Procedure for the preparation of divinyl derivatives 11a–d: The starting compound (250 mg, 0.96 mmol) was taken up in dry CH_2Cl_2 (15 mL) and the mixture was cooled to 0°C. Triethylamine (0.2 mL, 1.44 mmol) was added, followed by tosyl chloride (220 mg, 1.15 mmol), and the reaction mixture was stirred at 0°C for 4 h. After completion of the reaction, solvent was evaporated and the residue was dissolved in ether. The organic layer was washed with water, followed by brine, and dried on Na_2SO_4. Column chromatography over silica gel gave the tosylated product (350 g, 88%), which was stirred in a glass pressure bomb with allylamine (5 mL) in methanol (5 mL) at 65°C for 6 h. The reaction mixture was concentrated, and the residue was dissolved in THF. Triethylamine (1.0 mL) was added at 0°C, followed by benzyl chloroformate (170 mg, 0.99 mmol). The reaction mixture was stirred for 3 h. After completion of the reaction it was diluted with ethyl acetate and organic layer was washed with water, followed by brine, dried over anhydrous Na_2SO_4, and concentrated in vacuo. The residue was chromatographed over silica gel with ethyl acetate in hexane (25%) as eluent to furnish the pure divinyl product.

(2*R*,3*R*)-Allyl-(2-*tert*-butoxycarbonylamino-3-methoxymethoxy-pent-4-enyl)-carbamic acid benzyl ester (11a): Overall yield = 59%; $R_f=0.4$ (ethyl acetate in hexane, 20%); $[\alpha]_D^{25}=+9.2$ ($c=0.426$ in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): $\delta=7.34$ (s, 5H; Ar*H*), 5.82–5.66 (m, 2H), 5.31–5.12 (m, 4H), 5.15 (s, 2H), 4.67–4.52 (m, 2H), 4.21–4.05 (m, 2H), 3.91–3.88 (m, 1H), 3.79–3.71 (m, 1H), 3.39 (s, 3H; OCH_3), 3.35–3.26 (m, 2H), 1.44 ppm (s, 9H; OC(CH_3)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25°C, TMS): $\delta=155.8$ (C=O), 154.3 (C=O), 135.4, 133.7, 132.2, 127.2, 126.9, 126.6, 117.4, 115.6, 94.9, 93.3, 77.7, 77.1, 66.25, 65.9, 54.4, 51.6, 48.1, 44.4, 28.4 ppm; IR (neat): $\tilde{\nu}=3447$, 2932, 2362, 1704, 1640, 1464, 1243 cm^{-1}; MS (ESI): *m*/*z* (%): 457.1 (53) $[M+Na]^+$, 434.9 (50) $[M]^+$, 378.8 (42) $[M-t\text{Bu}]^+$, 335.2 (100) $[M-t\text{Boc}]^+$; elemental analysis (%) calcd for $C_{23}H_{34}N_2O_6$: C 63.57, H 7.89, N 6.45; found: C 63.55, H 7.68, N 6.25.

(2*R*,3*S*)-Allyl-(2-*tert*-butoxycarbonylamino-3-methoxymethoxy-pent-4-enyl)-carbamic acid benzyl ester (11b): Overall yield = 58%; $R_f=0.4$ (ethyl acetate in hexane, 20%); $[\alpha]_D^{25}=+11.2$ ($c=0.413$ in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25°C, TMS): $\delta=7.36$ (s, 5H; Ar*H*), 5.86–5.74 (m, 2H), 5.30–5.11 (m, 4H), 5.16 (s, 2H), 4.69–4.55 (m, 2H), 4.21–4.27 (m, 2H), 4.12–3.81 (m, 1H), 3.79–3.73 (m, 1H), 3.38 (s, 3H; OCH_3), 3.35–3.26 (m, 2H), 1.40 ppm (s, 9H; OC(CH_3)$_3$); MS (ESI): *m*/*z* (%): 457.1 (100) $[M+Na]^+$, 434.9 (50) $[M]^+$, 378.8 (40) $[M-t\text{Bu}]^+$, 335.2 (20) $[M-t\text{Boc}]^+$; elemental analysis (%) calcd for $C_{23}H_{34}N_2O_6$: C 63.57, H 7.89, N 6.45; found: C 63.48, H 7.79, N 6.47.

(2*S*,3*S*)-Allyl-(2-*tert*-butoxycarbonylamino-3-methoxymethoxy-pent-4-enyl)-carbamic acid benzyl ester (11c): Overall yield = 63%; $R_f=0.4$ (ethyl acetate in hexane, 20%); $[\alpha]_D^{25}=-10.7$ ($c=0.236$ in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): $\delta=7.38$ (s, 5H; Ar*H*), 5.78–5.70 (m, 2H), 5.36–5.08 (m, 4H), 5.14 (s, 2H), 4.67–4.51 (m, 2H), 4.21–4.04 (m, 2H), 3.94–3.79 (m, 1H), 3.78–3.74 (m, 1H), 3.39 (s, 3H; OCH_3), 3.35–3.26 (m, 2H), 1.44 ppm (s, 9H; OC(CH_3)$_3$); ^{13}C NMR (75 MHz, $CDCl_3$, 25°C, TMS): $\delta=155.9$, 154.6, 135.4, 133.5, 132.1, 127.1, 126.4, 117.4, 116.0, 115.5, 93.5, 77.8, 77.4, 66.2, 66.0, 54.4, 51.5, 51.2, 48.6, 48.1, 44.3, 43.5, 27.1 ppm; MS (ESI): *m*/*z* (%): 457.1 (53) $[M+Na]^+$, 378.8 (60) $[M-t\text{Bu}]^+$, 335.2 (100) $[M-t\text{Boc}]^+$; elemental analysis (%) calcd for $C_{23}H_{34}N_2O_6$: C 63.57, H 7.89, N 6.45; found: C 63.42, H 7.73, N 6.35.

(2*S*,3*R*)-Allyl-(2-*tert*-butoxycarbonylamino-3-methoxymethoxy-pent-4-enyl)-carbamic acid benzyl ester (11d): Overall yield = 60%; $R_f=0.41$ (ethyl acetate in hexane, 20%); $[\alpha]_D^{25}=-13.7$ ($c=0.209$ in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): $\delta=7.33$ (s, 5H; Ar*H*), 5.78–5.70 (m, 2H), 5.36–5.08 (m, 4H), 5.14 (s, 2H), 4.67–4.51 (m, 2H), 4.20–4.07 (m, 2H), 3.95–3.88 (m, 1H), 3.79–3.74 (m, 1H), 3.39 (s, 3H; OCH_3), 3.35–3.26 (m, 2H), 1.40 ppm (s, 9H; OC(CH_3)$_3$); ^{13}C NMR (75 MHz, $CDCl_3$, 25°C, TMS): $\delta=155.6$, 154.6, 135.4, 133.4, 133.3, 132.1, 127.1, 126.6, 126.4, 117.9, 117.4, 115.5, 93.3, 77.9, 77.1, 65.9, 54.4, 51.6, 48.1, 44.3, 28.4, 27.0 ppm; MS (ESI): *m*/*z* (%): 457.1 (40) $[M+Na]^+$, 434.9 (40) $[M]^+$, 378.8 (80) $[M-t\text{Bu}]^+$, 335.2 (100) $[M-t\text{Boc}]^+$; elemental analysis (%)

calcd for $C_{23}H_{34}N_2O_6$: C 63.57, H 7.89, N 6.45; found: C 63.39, H 7.81, N 6.37.

Genaral Procedure for the preparation of divinyl derivatives 12a–d: The starting compound was taken up in dry CH_2Cl_2 and cooled at 0 °C. Triethylamine was added, followed by tosyl chloride. The reaction mixture was stirred at the same temperature for 3 h. After completion of the reaction, solvent was evaporated, and the residue was dissolved in ether. The organic layer was washed with water, followed by brine, and dried on anhydrous Na_2SO_4. Column chromatography over silica gel gave the tosylated product, which was heated in a steel bomb with allylamine and methanol at 65 °C for 10–12 h. The reaction mixture was concentrated, and the residue was dissolved in CH_2Cl_2. Triethylamine was added at 0 °C, followed by TsCl. The reaction mixture was stirred for 3 h. After completion of the reaction the mixture was diluted with ethyl acetate, and the organic layer was washed with water, followed by brine, and dried over sodium sulfate. Chromatography over silica gel gave the divinyl product.

(3*R*,4*R*)-(1-{[Allyl-(toluene-4-sulfonyl)-amino]-methyl}-2-benzyloxy-but-3-enyl)-carbamic acid *tert*-butyl ester (12a): Overall yield = 65 %; R_f = 0.4 (ethyl acetate in hexane, 15 %); m.p. 105 °C; $[\alpha]_D^{25}$ = +6.3 (c = 1.850 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ = 7.69 (d, J = 8.1 Hz, 2H; Ar*H*), 7.37–7.23 (m, 7H; Ar*H*), 5.89–5.70 (m, 1H; 5-*H*), 5.67–5.48 (m, 1H; 8-*H*), 5.38–5.29 (m, 2H; 6-*H*), 5.19–5.11 (m, 2H; 9-*H*), 5.00 (d, J = 7.08 Hz, 1H; N*H*), 4.63 (d, J = 11.97 Hz, 1H; -C*H*$_2$Ph), 4.34 (d, J = 12.0 Hz, 1H; -C*H*$_2$Ph), 4.03 (pseudo t, 1H), 3.92–3.83 (m, 3H), 3.52 (dd, J_1 = 10.47 Hz, J_2 = 14.6 Hz, 1H), 3.15 (dd, J_1 = 4.68 Hz, J_2 = 14.6 Hz, 1H), 2.44 (s, 3H; C*H*$_3$), 1.45 ppm {s, 9H; -OC(C*H*$_3$)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): δ = 159 (C=O), 143, 139, 137, 135, 131, 130, 129, 127, 127.7, 120.1, 118.5, 81.3, 78, 71, 51, 50, 46, 30, 27.7, 21.8 ppm; IR (neat): $\tilde{\nu}$ = 3953.1, 3906.5, 3755.2, 3375.0, 2980.3, 2932.4, 2369.0, 1690.7, 1597.5, 1534.4, 1352.8, 1159.5, 1091.4, 1046.1, 988.7, 921.8, 808.5, 759.0, 704.0, 661.8, 550.5 cm^{-1}; MS (FAB): m/z (%): 501 (15) [M+H]$^+$, 445 (30) [M−tBu]$^+$, 401 (100) [M−tBoc]$^+$; elemental analysis (%) calcd for $C_{27}H_{36}N_2O_5S$: C 64.77, H 7.25, N 5.60; found: C 64.52, H 7.20, N 5.44.

(3*R*,4*S*)-(1-{[Allyl-(toluene-4-sulfonyl)-amino]-methyl}-2-benzyloxy-but-3-enyl)-carbamic acid *tert*-butyl ester (12b): Overall yield = 64 %; R_f = 0.4 (ethyl acetate in hexane, 15 %); m.p. 112 °C; $[\alpha]_D^{25}$ = +11.23 (c = 0.824 in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): δ = 7.66 (d, J = 8.1 Hz, 2H; Ar*H*), 7.32–7.24 (m, 7H; Ar*H*), 5.89–5.68 (m, 1H; 5-*H*), 5.61–5.40 (m, 1H; 8-*H*), 5.37–5.26 (m, 2H; 6-*H*), 5.18–5.08 (m, 2H; 9-*H*), 4.95 (d, J = 6.2 Hz, 1H; N*H*), 4.61 (d, J = 11.8 Hz, 1H; -C*H*$_2$Ph), 4.34 (d, J = 11.8 Hz, 1H; -C*H*$_2$Ph), 4.0 (pseudo t, J = 4.5 Hz, 1H; 3-*H*), 3.82–3.76 (m, 3H), 3.51–3.39 (m, 1H), 3.18–3.08 (m, 1H), 2.43 (s, 3H; CH$_3$), 1.42 ppm (s, 9H; -OC(C*H*$_3$)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): δ = 156 (C=O), 143, 138, 137, 135, 132, 130, 129, 128, 127.7, 120.1, 119, 81.3, 79, 71, 52, 51, 46, 30, 28, 21.9 ppm; IR (neat): $\tilde{\nu}$ = 3377, 3315, 3247, 2812, 2374, 1596, 1350, 1163, 763, 663, 553 cm^{-1}; MS (FAB): m/z (%): 501 (15) [M+H]$^+$, 445 (30) [M−tBu]$^+$, 401 (100) [M−tBoc]$^+$; elemental analysis (%) calcd for $C_{27}H_{36}N_2O_5S$: C 64.77, H 7.25, N 5.60; found: C 64.68, H 7.18, N 5.53.

(3*S*,4*S*)-(1-{[Allyl-(toluene-4-sulfonyl)-amino]-methyl}-2-benzyloxy-but-3-enyl)-carbamic acid *tert*-butyl ester (12c): Overall yield = 69 %; R_f = 0.41 (ethyl acetate in hexane, 15 %); m.p. 102 °C; $[\alpha]_D^{25}$ = −6.76 (c = 0.700 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ = 7.68 (d, J = 8.3 Hz, 2H; Ar*H*), 7.39–7.28 (m, 7H; Ar*H*), 5.86–5.71 (m, 1H; 5-*H*), 5.61–5.49 (m, 1H; 8-*H*), 5.38–5.29 (m, 2H; 6-*H*), 5.19–5.11 (m, 2H; 9-*H*), 5.00 (d, J = 8.67 Hz, 1H; N*H*), 4.62 (d, J = 11.8 Hz, 1H; -C*H*$_2$Ph), 4.36 (d, J = 11.8 Hz, 1H; -C*H*$_2$Ph), 4.04 (pseudo t, J = 4.6 Hz, 1H; 3-*H*), 3.92–3.80 (m, 3H), 3.51 (dd, J_1 = 10.5 Hz, J_2 = 14.5 Hz, 1H), 3.16 (dd, J_1 = 3.84 Hz, J_2 = 14.6 Hz, 1H), 2.44 (s, 3H; CH$_3$), 1.42 ppm (s, 9H; -OC(C*H*$_3$)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): δ = 156.2 (C=O), 143.6, 138.6, 137.5, 135.4, 133, 130.1, 128.7, 128.0, 127.6, 119.9, 119.2, 81.3, 79.7, 74.4, 71.3, 52.4, 51.1, 46.2, 28.8, 21.9 ppm; IR (neat): $\tilde{\nu}$ = 3960.1, 3907.0, 3859.8, 3372.9, 2369.4, 1595.9, 1352.0, 1159.3, 1091.9 cm^{-1}; MS (ESI): m/z (%): 523.1 (100) [M+Na]$^+$, 445.1 (30) [M−tBu]$^+$, 401.1 (50) [M−tBoc]$^+$; elemental analysis (%) calcd for $C_{27}H_{36}N_2O_5S$: C 64.77, H 7.25, N 5.60; found: C 64.71, H 7.20, N 5.52.

(3*S*,4*R*)-(1-{[Allyl-(toluene-4-sulfonyl)-amino]-methyl}-2-benzyloxy-but-3-enyl)-carbamic acid *tert*-butyl ester (12d): Overall yield = 61 %; R_f = 0.4 (ethyl acetate in hexane, 15 %); $[\alpha]_D^{25}$ = −10.23 (c = 0.727 in $CHCl_3$); m.p. 115 °C; ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ = 7.69 (d, J = 8.3 Hz, 2H; Ar*H*), 7.37–7.37 (m, 7H; Ar*H*), 5.81–5.71 (m, 1H), 5.62–5.48 (m, 1H), 5.38–5.28 (m, 2H), 5.19–5.11 (m, 2H), 5.01 (d, J = 8.7 Hz, 1H), 4.63 (d, J = 11.9 Hz, 1H), 4.37 (d, J = 11.9 Hz, 1H), 4.03 (pseudo t, J = 4.3 Hz, 1H), 3.92–3.80 (m, 3H), 3.51 (dd, J_1 = 10.5 Hz, J_2 = 14.6 Hz, 1H), 3.16 (dd, J_1 = 3.84 Hz, J_2 = 14.6 Hz, 1H), 2.44 (s, 3H; C*H*$_3$), 1.43 ppm (s, 9H; OC(C*H*$_3$)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): δ = 156.2, 143.6, 138.6, 137.5, 135.4, 132.9, 130.1, 128.7, 128.0, 127.6, 119.9, 119.2, 81.3, 79.7, 71.3, 52.4, 51.0, 46.2, 28.8, 21.9 ppm; MS (ESI): m/z (%): 523.1 (100) [M+Na]$^+$, 445.1 (40) [M−tBu]$^+$, 401.1 (50) [M−tBoc]$^+$; elemental analysis (%) calcd for $C_{27}H_{36}N_2O_5S$: C 64.77, H 7.25, N 5.60; found: C 64.62, H 7.19, N 5.54.

General Procedure for RCM: Grubbs' catalyst (5 mol %) was added to a solution of the starting compound at reflux in dry CH_2Cl_2, and the reaction mixture was stirred overnight. After completion of the reaction, solvent was removed and the residue was chromatographed over silica gel to furnish the tetrahydroazepine core.

(3*R*,4*R*)-3-*tert*-Butoxycarbonylamino-4-methoxymethoxy-2,3,4,7-tetrahydro-azepine-1-carboxylic acid benzyl ester (9a): Yield = 76 %; R_f = 0.4 (ethyl acetate in hexane, 30 %); $[\alpha]_D^{25}$ = −40.5 (c = 2.1 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ = 7.35 (s, 5H; Ar*H*), 5.98–5.89 (m, 1H), 5.82–5.72 (m, 1H), 5.15 (s, 2H), 4.65–4.54 (m, 2H), 4.35 (d, J = 12.6 Hz, 1H), 4.20 (dd, J = 17.0, 3.6 Hz, 2H), 3.91–3.65 (m, 3H), 3.32 (s, 3H; OC*H*$_3$), 1.43 ppm (s, 9H; OC(C*H*$_3$)$_3$); ^{13}C NMR (75 MHz, $CDCl_3$, 25 °C, TMS): δ = 154.5, 149.3 (C=O), 129.6, 127.2, 126.7, 126.5, 94.9, 93.9, 78.14, 73.6, 66.1, 54.2, 52.0, 50.2, 47.9, 45.3, 28.42 ppm; IR (neat): $\tilde{\nu}$ = 3860, 3445, 2932, 2362, 1700, 1636, 1387, 1167, 1099, 1033 cm^{-1}; MS (ESI): m/z (%): 429.1 (100) [M+Na]$^+$, 407.0 (10) [M]$^+$, 373.1 (12) [M−tBu+Na]$^+$, 307.1 (13) [M−tBoc]$^+$; elemental analysis (%) calcd for $C_{21}H_{30}N_2O_6$: C 62.05, H 7.44, N 6.89; found: C 61.97, H 7.44, N 6.78.

(3*R*,4*S*)-3-*tert*-Butoxycarbonylamino-4-methoxymethoxy-2,3,4,7-tetrahydro-azepine-1-carboxylic acid benzyl ester (9b): Yield = 78.4 %; R_f = 0.4 (ethyl acetate in hexane, 30 %); $[\alpha]_D^{25}$ = −34.5 (c = 2.3 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ = 7.38 (s, 5H; Ar*H*), 5.93 (pseudo t, 1H), 5.79–5.77 (m, 1H), 5.15 (s, 2H), 4.71–4.58 (m, 2H), 4.36 (d, J = 12.6 Hz, 1H), 4.22 (dd, J = 17.0, 3.6 Hz, 2H), 3.86–3.69 (m, 3H), 3.35 (s, 3H; OC*H*$_3$), 1.43 ppm (s, 9H; OC(C*H*$_3$)$_3$); ^{13}C NMR (75 MHz, $CDCl_3$, 25 °C, TMS): δ = 154.5, 135.3, 129.6, 127.2, 126.7, 125.6, 94.0, 78.2, 73.5, 66.1, 63.9, 54.2, 50.2, 47.9, 45.3, 30.6, 28.4, 27.0, 21.3 ppm; MS (ESI): m/z (%): 429.1 (100) [M+Na]$^+$, 407.0 (20) [M]$^+$, 350.1 (10) [M−tBu]$^+$, 307.1 (80) [M−tBoc]$^+$; elemental analysis (%) calcd for $C_{21}H_{30}N_2O_6$: C 62.05, H 7.44, N 6.89; found: C 62.01, H 7.29, N 6.79.

(3*S*,4*S*)-3-*tert*-Butoxycarbonylamino-4-methoxymethoxy-2,3,4,7-tetrahydro-azepine-1-carboxylic acid benzyl ester (9c): Yield = 78 %; R_f = 0.41 (ethyl acetate in hexane, 30 %); $[\alpha]_D^{25}$ = +48.5 (c = 2.1 in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): δ = 7.36 (s, 5H; Ar*H*), 5.89 (br s, 1H), 5.72 (br s, 1H), 5.23 (s, 2H), 4.69–4.49 (m, 2H), 4.31 (br s, 1H), 4.08 (br s, 2H), 3.82–3.61 (m, 3H), 3.24 (s, 3H; OC*H*$_3$), 1.35 ppm (s, 9H; OC(C*H*$_3$)$_3$); MS (ESI): m/z (%): 429.1 (100) [M+Na]$^+$, 407.0 (10) [M]$^+$, 373.1 (15) [M−tBu+Na]$^+$, 307.1 (13) [M−tBoc]$^+$; elemental analysis (%) calcd for $C_{21}H_{30}N_2O_6$: C 62.05, H 7.44, N 6.89; found: C 61.89, H 7.19, N 6.75.

(3*S*,4*R*)-3-*tert*-Butoxycarbonylamino-4-methoxymethoxy-2,3,4,7-tetrahydro-azepine-1-carboxylic acid benzyl ester (9d): Yield = 78.5 %; R_f = 0.41 (ethyl acetate in hexane, 30 %); $[\alpha]_D^{25}$ = +39.5 (c = 2.2 in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): δ = 7.36 (s, 5H; Ar*H*), 5.94–5.92 (m, 1H), 5.79–5.74 (m, 1H), 5.16 (s, 2H), 4.67–4.58 (m, 2H), 4.39 (br s, 1H), 4.20 (d, J = 8.6 Hz, 2H), 3.91–3.69 (m, 3H), 3.32 (s, 3H; OC*H*$_3$), 1.43 ppm (s, 9H; OC(C*H*$_3$)$_3$); ^{13}C NMR (75 MHz, $CDCl_3$, 25 °C, TMS): δ = 154.5, 136.4, 130.1, 129.6, 127.2, 126.7, 94.0, 78.2, 73.5, 66.3, 66.1, 54.3, 50.1, 47.9, 45.3, 28.4, 27.0 ppm; MS (ESI): m/z (%): 429.1 (100) [M+Na]$^+$, 407.0 (10) [M]$^+$, 373.1 (35) [M−tBu+Na]$^+$, 307.1 (13) [M−tBoc]$^+$; elemental analysis (%) calcd for $C_{21}H_{30}N_2O_6$: C 62.05, H 7.44, N 6.89; found: C 61.91, H 7.23, N 6.81.

(3*R*,4*R*)-[4-Benzyloxy-1-(toluene-4-sulfonyl)-2,3,4,7-tetrahydro-1*H*-azepin-3-yl]-carbamic acid *tert*-butyl ester (10a): Yield = 76%; R_f = 0.4 (ethyl acetate in hexane, 25%); $[\alpha]_D^{25}$ = +7.14 (c = 0.370 in $CHCl_3$); ^{1}H NMR ($CDCl_3$, 300 MHz, 25°C, TMS): δ = 7.63 (d, J = 8.0 Hz, 2H; Ar*H*), 7.35–7.23 (m, 7H; Ar*H*), 5.80–5.68 (m, 2H; 5-*H*, 6-*H*), 4.43 (dd, J_1 = 12 Hz, J_2 = 22 Hz, 2H; -C*H*$_2$Ph), 4.37 (brs, 1H), 4.29 (brs, 1H; 4-*H*), 3.76 (brs, 2H; 2-*H*, 7-*H*), 3.48–3.32 (m, 2H; 2′-*H*, 7′-*H*), 2.38 (s, 3H; C*H*$_3$), 1.48 ppm (s, 9H; -OC(C*H*$_3$)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$): δ = 156.1 (C=O), 143.9, 138.2, 136.1, 133.4, 130.2, 128.8, 128.1, 127.6, 80.1, 71.2, 51.9, 50.2, 48.4, 32.3, 30.1, 28.8, 27.1, 23.1, 21.9, 14.5 ppm; IR (neat): $\tilde{\nu}$ = 3980, 3950, 3711, 1838, 1716, 1500, 1365, 1163, 764, 673 cm^{-1}; MS (FAB): m/z (%): 473 (15) [M+H]$^+$, 417 (70) [M−*t*Bu]$^+$, 373 (90) [M−*t*Boc]$^+$, 91 (100) [PhCH$_2$]$^+$; elemental analysis (%) calcd for $C_{25}H_{32}N_2O_5S$: C 63.54, H 6.82, N 5.93; found: C 63.51, H 6.73, N 5.85.

(3*R*,4*S*)-[4-Benzyloxy-1-(toluene-4-sulfonyl)-2,3,4,7-tetrahydro-1*H*-azepin-3-yl]-carbamic acid *tert*-butyl ester (10b): Yield = 79%; R_f = 0.42 (ethyl acetate in hexane, 25%); $[\alpha]_D^{25}$ = +4.21 (c = 0.190 in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25°C, TMS): δ = 7.64 (d, 2H; J = 8.2 Hz, Ar*H*), 7.28–7.20 (m, 7H; Ar*H*), 5.96–5.71 (m, 2H; 5-*H*, 6-*H*), 4.82 (d, J = 8 Hz, 1H; N*H*), 4.52 (dd, J_1 = 11.2 Hz, J_2 = 22.0 Hz, 2H; -C*H*$_2$Ph), 4.39–4.29 (m, 2H; 3-*H*, 4-*H*), 3.77 (m, 2H; 2-*H*, 7-*H*), 3.61–3.34 (m, 2H; 2′-*H*, 7′-*H*), 2.41 (s, 3H; C*H*$_3$), 1.43 ppm (s, 9H; -OC(C*H*$_3$)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25°C, TMS): δ = 155.8 (C=O), 143.5, 138.2, 136.5, 133.4, 130.1, 128.7, 128.1, 127.6, 96.6, 79.8, 71.1, 51.8, 50.2, 48.4, 28.8, 21.9 ppm; IR (neat): $\tilde{\nu}$ = 3980, 3950, 3711, 1838, 1716, 1500, 1365, 1163, 764, 673 cm^{-1}; MS (FAB): m/z (%): 473 (65) [M+H]$^+$, 417 (70) [M−*t*Bu]$^+$, 373 (90) [M−*t*Boc]$^+$, 91 (100) [PhCH$_2$]$^+$; elemental analysis (%) calcd for $C_{25}H_{32}N_2O_5S$: C 63.54, H 6.82, N 5.93; found: C 63.45, H 6.78, N 5.79.

(3*S*,4*S*)-[4-Benzyloxy-1-(toluene-4-sulfonyl)-2,3,4,7-tetrahydro-1*H*-azepin-3-yl]-carbamic acid *tert*-butyl ester (10c): Yield = 82%; R_f = 0.4 (ethyl acetate in hexane, 25%); $[\alpha]_D^{25}$ = −8.20 (c = 0.256 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): δ = 7.67 (d, J = 8.2 Hz, 2H; Ar*H*), 7.38–7.28 (m, 7H; Ar*H*), 5.85–5.73 (m, 2H; 5-*H*, 6-*H*), 4.91 (d, J = 8.4 Hz, 1H; N*H*), 4.56 (dd, J_1 = 11.7 Hz, J_2 = 25.0 Hz, 2H), 4.43 (s, 1H), 4.35 (brs, 1H), 3.79 (d, J = 3.3 Hz, 2H), 3.56–3.42 (m, 2H), 2.43 (s, 3H; C*H*$_3$), 1.47 ppm (s, 9H; -OC(C*H*$_3$)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25°C, TMS): δ = 156.1 (C=O), 143.9, 138.2, 136.1, 133.4, 130.2, 128.8, 128.1, 127.6, 80.1, 73.8, 73.1, 71.2, 51.9, 50.2, 48.4, 28.7, 21.9 ppm; MS (FAB): m/z (%): 473 (45) [M+Na]$^+$, 417 (100) [M−*t*Bu]$^+$, 373 (90) [M−*t*Boc]$^+$; elemental analysis (%) calcd for $C_{25}H_{32}N_2O_5S$: C 63.54, H 6.82, N 5.93; found: C 63.481, H 6.81, N 5.89.

(3*S*,4*R*)-[4-Benzyloxy-1-(toluene-4-sulfonyl)-2,3,4,7-tetrahydro-1*H*-azepin-3-yl]-carbamic acid *tert*-butyl ester (10d): Yield = 75%; R_f = 0.4 (ethyl acetate in hexane, 25%); $[\alpha]_D^{25}$ = −5.22 (c = 0.134 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): δ = 7.61 (d, J = 8 Hz, 2H; Ar*H*), 7.32–7.22 (m, 7H; Ar*H*), 5.89–5.68 (m, 2H; 5-*H*, 6-*H*), 4.86 (d, J = 9.0 Hz, 1H; N*H*), 4.52 (dd, J_1 = 12.3 Hz, J_2 = 21 Hz, 2H; -CH$_2$Ph), 4.37 (brs, 1H; 3-*H*), 4.30 (brs, 1H; 4-*H*), 3.76 (brs, 2H; 2-*H*, 7-*H*), 3.47–3.41 (m, 2H; 2′-*H*, 7′-*H*), 2.38 (s, 3H; C*H*$_3$), 1.41 ppm (s, 9H; -OC(C*H*$_3$)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25°C, TMS): δ = 143.9 (C=O), 138.1, 136.0, 133.45, 130.2, 128.8, 128.1, 127.56, 80.07, 71.2, 51.9, 50.2, 48.4, 30.1, 28.7, 21.9 ppm; MS (ESI): m/z (%): 496.2 (100) [M+Na]$^+$, 417.1 (70) [M−*t*Bu]$^+$, 373 (90) [M−*t*Boc]$^+$; elemental analysis (%) calcd for $C_{25}H_{32}N_2O_5S$: C 63.54, H 6.82, N 5.93; found: C 63.49, H 6.72, N 5.84.

General Procedure for hydrogenation to prepare 21a–d: The starting compound (500 mg) was taken up in THF (25 mL). Pd/C (10%, 25 mg) was added, and the reaction mixture was stirred under hydrogen at room temperature. The solution was filtered through Celite, and the filtrate was concentrated and passed through a small silica column to furnish the product.

(3*R*,4*R*)-3-*tert*-Butoxycarbonylamino-4-methoxymethoxy-azepane-1-carboxylic acid benzyl ester (21a): Yield = 95%; R_f = 0.42 (ethyl acetate in hexane, 30%); $[\alpha]_D^{25}$ = +19.5 (c = 0.496 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): δ = 7.44–7.30 (m, 5H; Ar*H*), 5.26–5.11 (m, 1H), 5.17 (s, 2H), 4.65 (dd, J = 12.6, 6.6 Hz, 2H), 3.94–3.88 (m, 2H), 3.75–3.67 (m, 2H), 3.43 (s, 3H; OC*H*$_3$), 3.33–3.24 (m, 2H), 2.07–1.67 (m, 4H), 1.46 ppm (s, 9H; OC(C*H*$_3$)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25°C, TMS): δ = 156.5 (C=O), 155.4 (C=O), 137.4, 128.8, 128.3, 96.4, 79.8, 67.6, 56.3, 53.9, 47.6, 47.2, 30.1, 28.8, 27.6, 20.6 ppm; IR (neat): $\tilde{\nu}$ = 3861, 3754, 3445, 3021, 2364, 1702, 1217, 765 cm^{-1}; MS (ESI): m/z (%): 431.2 (100) [M+Na]$^+$, 399.2 (20) [M−OCH$_3$+Na]$^+$, 375.2 (12) [M−*t*Bu+Na]$^+$, 309.2 (13) [M−*t*Boc]$^+$; elemental analysis (%) calcd for $C_{21}H_{32}N_2O_6$: C 61.75, H 7.90, N 6.86; found: C 61.64, H 7.88, N 6.82.

(3*R*,4*S*)-3-*tert*-Butoxycarbonylamino-4-methoxymethoxy-azepane-1-carboxylic acid benzyl ester (21b): Yield = 93%; R_f = 0.41 (ethyl acetate in hexane, 30%); $[\alpha]_D^{25}$ = +26.5 (c = 0.269 in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25°C, TMS): δ = 7.45 (s, 5H; Ar*H*), 5.24–5.09 (m, 1H), 5.16 (s, 2H), 4.66 (dd, J = 12.8, 6.4 Hz, 2H), 3.94–3.88 (m, 2H), 3.75–3.65 (m, 2H), 3.43 (s, 3H; OC*H*$_3$), 3.33–3.24 (m, 2H), 2.07–1.66 (m, 4H), 1.45 ppm (s, 9H; OC(C*H*$_3$)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25°C, TMS): δ = 156.5, 155.3, 137.2, 128.8, 128.2, 96.6, 96.3, 79.7, 67.6, 67.4, 56.3, 53.8, 47.6, 47.1, 30.0, 28.1, 27.7, 21.6 ppm; MS (ESI): m/z (%): 431.2 (100) [M+Na]$^+$, 399.2 (20) [M−OCH$_3$+Na]$^+$, 375.2 (12) [M−*t*Bu+Na]$^+$, 309.2 (13) [M−*t*Boc]$^+$; elemental analysis (%) calcd for $C_{21}H_{32}N_2O_6$: C 61.75, H 7.90, N 6.86; found: C 61.71, H 7.86, N 6.76.

(3*S*,4*S*)-3-*tert*-Butoxycarbonylamino-4-methoxymethoxy-azepane-1-carboxylic acid benzyl ester (21c): Yield = 95%; R_f = 0.41 (ethyl acetate in hexane, 30%); $[\alpha]_D^{25}$ = −15.5 (c = 0.396 in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25°C, TMS): δ = 7.42–7.26 (m, 5H; Ar*H*), 5.26–5.15 (m, 1H), 5.14 (s, 2H), 4.66 (dd, J = 12.6, 6.6 Hz, 2H), 3.94–3.67 (m, 4H), 3.39 (s, 3H; OC*H*$_3$), 3.33–3.27 (m, 2H), 2.07–1.67 (m, 4H), 1.44 ppm (s, 9H; OC(C*H*$_3$)$_3$); MS (ESI): m/z (%): 431.2 (100) [M+Na]$^+$, 399.2 (20) [M−OCH$_3$+Na]$^+$, 375.2 (50) [M−*t*Bu+Na]$^+$, 309.2 (13) [M−*t*Boc]$^+$; elemental analysis (%) calcd for $C_{21}H_{32}N_2O_6$: C 61.75, H 7.90, N 6.86; found: C 61.71, H 7.79, N 6.80.

(3*S*,4*R*)-3-*tert*-Butoxycarbonylamino-4-methoxymethoxy-azepane-1-carboxylic acid benzyl ester (21d): Yield = 96%; R_f = 0.42 (ethyl acetate in hexane, 30%); $[\alpha]_D^{25}$ = −29.5 (c = 0.278 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): δ = 7.48–7.29 (m, 5H; Ar*H*), 5.28–5.12 (m, 1H), 5.17 (s, 2H), 4.65 (dd, J = 12.6, 6.6 Hz, 2H), 3.92–3.90 (m, 2H), 3.76–3.66 (m, 2H), 3.43 (s, 3H; OC*H*$_3$), 3.42–3.29 (m, 2H), 2.08–1.69 (m, 4H), 1.44 ppm (s, 9H; OC(C*H*$_3$)$_3$); ^{13}C NMR (50 MHz, $CDCl_3$, 25°C, TMS): δ = 156.5, 155.3, 137.3, 128.8, 128.2, 96.3, 79.7, 67.5, 56.3, 53.8, 47.6, 47.2, 30.1, 28.7, 27.7, 20.6 ppm; MS (ESI): m/z (%): 431.2 (100) [M+Na]$^+$, 399.2 (35) [M−OCH$_3$+Na]$^+$, 352.2 (55) [M−*t*Bu]$^+$, 309.2 (13) [M−*t*Boc]$^+$; elemental analysis (%) calcd for $C_{21}H_{32}N_2O_6$: C 61.75, H 7.90, N 6.86; found: C 61.71, H 7.82, N 6.79.

General Procedure for preparation of amido alcohols 22a–d: The starting compound was stirred with TFA (50%) at room temperature for 30 min. The solvent was evaporated, and coevaporation was performed with dry CH_2Cl_2 to remove excess TFA. The residue was dissolved in dry CH_2Cl_2, and triethylamine was added at 0°C, followed by *p*-benzyloxybenzoyl chloride. The reaction mixture was stirred at room temperature for 1 h. Solvent was evaporated, and the residue was dissolved in ethyl acetate. The organic layer was washed with $NaHCO_3$ (5%), followed by brine, and dried over sodium sulfate. Chromatography over silica gel furnished the amido alcohols.

(3*R*,4*R*)-3-(4-Benzyloxybenzoylamino)-4-hydroxyazepane-1-carboxylic acid benzyl ester (22a): Yield = 59%; R_f = 0.3 (ethyl acetate in hexane, 50%); $[\alpha]_D^{25}$ = −82.85 (c = 2.1 in methanol); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): δ = 8.18 (d, J = 8.7 Hz, 1H), 7.80 (d, J = 7.98 Hz, 2H; Ar*H*), 7.47–7.31 (m, 10H; Ar*H*), 7.01 (d, J = 8.19 Hz, 2H; Ar*H*), 5.31 (s, 1H), 5.27–5.19 (m, 2H), 5.15 (s, 2H), 4.45 (s, 1H), 4.22 (d, J = 14.3 Hz, 1H), 4.04–3.94 (m, 2H), 3.19 (d, J = 14.5 Hz, 1H), 3.10–3.04 (m, 1H), 2.05–1.64 ppm (m, 4H); ^{13}C NMR (50 MHz, $CDCl_3$, 25°C, TMS): δ = 156.4, 139.3 135.1, 128.0, 127.4, 127.3, 126.9, 126.5, 126.1, 113.4, 68.81, 66.5, 46.2, 30.6, 28.4, 28.1, 27.7, 21.4 ppm; IR (neat): $\tilde{\nu}$ = 3906.5, 3867, 3820, 3440, 2924, 2857, 2363, 1636, 1427, 1380, 670 cm^{-1}; MS (FAB): m/z (%): 497.6 (100) [M+Na]$^+$; elemental analysis (%) calcd for $C_{28}H_{30}N_2O_5$: C 70.87, H 6.37, N 5.90; found: C 70.85, H 6.34, N 5.82.

(3*R*,4*S*)-3-(4-Benzyloxybenzoylamino)-4-hydroxyazepane-1-carboxylic acid benzyl ester (22b): Yield = 70%; R_f = 0.31 (ethyl acetate in hexane, 50%); $[\alpha]_D^{25}$ = −73.6 (c = 0.85 in methanol); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): δ = 7.20–6.99 (m, 14H; Ar*H*), 5.26–5.14 (m, 4H), 4.45 (s, 1H), 4.33–3.85 (m, 3H), 3.67–3.09 (m, 2H), 3.19 (d, J = 14.5 Hz, 1H), 3.10–3.04 (m, 1H), 2.05–1.62 ppm (m, 4H); ^{13}C NMR (75 MHz, $CDCl_3$,

25 °C, TMS): δ = 157.1, 135.1, 131.1, 130.6, 128.0, 127.4, 127.3, 127.0, 126.5, 126.2, 120.9, 113.5, 72.5, 68.9, 66.5, 55.4, 46.1, 30.6, 28.4, 21.4, 20.5, 12.8 ppm; MS (ESI): *m*/*z* (%): 497.2 (100) $[M+Na]^+$; elemental analysis (%) calcd for $C_{28}H_{30}N_2O_5$: C 70.87, H 6.37, N 5.90; found: C 70.34, H 6.14, N 5.59.

(3*S*,4*S*)-3-(4-Benzyloxybenzoylamino)-4-hydroxyazepane-1-carboxylic acid benzyl ester (22c): Yield = 67 %; R_f = 0.32 (ethyl acetate in hexane, 50 %); $[\alpha]_D^{25}$ = +79.6 (*c* = 1.25 in methanol); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ = 8.20 (d, *J* = 8.7 Hz, 1H), 7.80 (d, *J* = 7.98 Hz, 2H; Ar*H*), 7.47–7.31 (m, 10H; Ar*H*), 7.01 (d, *J* = 8.19 Hz, 2H; Ar*H*), 5.30–5.12 (m, 4H), 4.43 (s, 1H), 4.17 (d, *J* = 14.3 Hz, 1H), 4.01–3.92 (m, 2H), 3.19 (d, *J* = 14.5 Hz, 1H), 3.10–3.04 (m, 1H), 2.05–1.60 ppm (m, 4H); ^{13}C NMR (75 MHz, $CDCl_3$, 25 °C, TMS): δ = 156.2, 135.0, 128.0, 127.4, 127.3, 126.9, 126.5, 126.1, 113.4, 72.3, 68.8, 66.3, 55.3, 46.1, 30.4, 28.4, 21.4, 19.1, 12.9 ppm; MS (ESI): *m*/*z* (%): 497.2 (100) $[M+Na]^+$; elemental analysis (%) calcd for $C_{28}H_{30}N_2O_5$: C 70.87, H 6.37, N 5.90; found: C 70.73, H 6.19, N 5.63.

(3*S*,4*R*)-3-(4-Benzyloxybenzoylamino)-4-hydroxyazepane-1-carboxylic acid benzyl ester (22d): Yield = 65 %; R_f = 0.3 (ethyl acetate in hexane, 50 %); $[\alpha]_D^{25}$ = +63.65 (*c* = 1.25 in methanol); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ = 7.81 (d, *J* = 7.98 Hz, 2H), 7.44–7.28 (m, 10H; Ar*H*), 7.01 (d, *J* = 8.19 Hz, 2H; Ar*H*), 5.30–5.12 (m, 4H), 4.45 (s, 1H), 4.20 (d, *J* = 14.3 Hz, 1H), 4.04–3.94 (m, 2H), 3.18 (d, *J* = 14.5 Hz, 1H), 3.10–3.04 (m, 1H), 2.05–1.63 ppm (m, 4H); ^{13}C NMR (75 MHz, $CDCl_3$, 25 °C, TMS): δ = 153.6, 138.4, 135.1, 128.0, 127.4, 127.3, 126.9, 126.4, 126.1, 113.4, 68.8, 66.2, 47.8, 46.1, 30.6, 28.4, 27.6, 21.4, 12.8 ppm; MS (ESI): *m*/*z* (%): 497.1 (100) $[M+Na]^+$; elemental analysis (%) calcd for $C_{28}H_{30}N_2O_5$: C 70.87, H 6.37, N 5.90; found: C 70.80, H 6.27, N 5.69.

General Procedure for preparation of amido alcohols 24a–d: The starting compound (200 mg, 0.424 mmol) was taken up in methanol and a catalytic amount of HCl was added, followed by Pd/C (10 %, 5 % *w*/*w*). The reaction mixture was stirred under hydrogen in a Parr apparatus at 40 psi pressure. The reaction mixture was filtered through Celite, and the filtrate was concentrated and dissolved in dry CH_2Cl_2 (10 mL). Triethylamine (0.5 mL) was added at 0 °C, followed by *p*-benzyloxybenzoyl chloride (170 mg, 0.70 mmol). The reaction mixture was stirred at room temperature for 1 h. Solvent was evaporated, and the residue was dissolved in ethyl acetate. The organic layer was washed with $NaHCO_3$ (5 %), followed by brine, and dried over sodium sulfate. Chromatography over silica gel furnished the amido alcohol.

(3*R*,4*R*)-4-Benzyloxy-*N*-[4-hydroxy-1-(toluene-4-sulfonyl)-azepan-3-yl]-benzamide (24a): Yield = 73 %; R_f = 0.3 (ethyl acetate in hexane, 60 %); m.p. 147 °C; $[\alpha]_D^{25}$ = +11.65 (*c* = 0.156 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ = 7.97 (d, *J* = 8.8 Hz, 2H; Ar*H*), 7.70 (d, *J* = 8.8 Hz, 2H; Ar*H*), 7.51–7.32 (m, 7H; Ar*H*), 7.05 (d, *J* = 8.8 Hz, 2H; Ar*H*), 5.15 (s, 2H; -CH_2Ph), 4.90 (br s, 1H; N*H*), 4.49 (s, 1H; 3-*H*), 3.89–3.71 (m, 3H; 2-*H*, 7-*H*, 4-*H*), 2.96–2.88 (m, 2H; 2′-*H*, 7′-*H*), 2.46 (s, 3H; CH_3), 2.06–1.95 (m, 6-*H*), 1.86–1.83 (m, 1H; 5-*H*), 1.65–1.51 ppm (m, 1H; 5′-H); ^{13}C NMR (75 MHz, $CDCl_3$, 25 °C, TMS): δ = 168.4, 160.56, 142.78, 135.13, 133.46, 128.69, 128.24, 127.36, 126.84, 126.16, 125.87, 124.46, 113.52, 68.83, 52.01, 47.29, 46.40, 27.24, 21.17, 20.22 ppm; IR (neat): $\tilde{\nu}$ = 3907, 3855, 3804, 3756, 3453, 2933, 2370, 1602, 1347, 1249, 1153, 1092, 756, 660 cm^{-1}; MS (ESI): *m*/*z* (%): 517.3 (35) $[M+Na]^+$, 495.2 (100) $[M+H]^+$; elemental analysis (%) calcd for $C_{27}H_{30}N_2O_5S$: C 65.57, H 6.11, N 5.66; found: C 65.37, H 6.04, N 5.64.

(3*R*,4*S*)-4-Benzyloxy-*N*-[4-hydroxy-1-(toluene-4-sulfonyl)-azepan-3-yl]-benzamide (24b): Yield = 71 %; R_f = 0.3 (ethyl acetate in hexane, 60 %); m.p. 142 °C; $[\alpha]_D^{25}$ = +3.65 (*c* = 0.245 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ = 7.92 (d, *J* = 8.8 Hz, 2H; Ar*H*), 7.69 (d, *J* = 8.8 Hz, 2H; Ar*H*), 7.47–7.26 (m, 7H; Ar*H*), 7.05 (d, *J* = 8.8 Hz, 2H; Ar*H*), 5.12 (s, 2H; CH_2Ph), 4.45 (s, 1H; N*H*), 4.00 (s, 1H; 3-*H*), 3.88–3.67 (m, 3H; 4-*H*, 2-*H*,7-*H*), 2.96–2.88 (m, 2H; 2′-*H*, 7′-*H*), 2.46 (s, 3H; CH_3), 2.06–1.66 ppm (m, 4H; 5-*H*, 6-*H*); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): δ = 169.8, 161.8, 144.1, 136.3, 134.5, 130.0, 129.5, 129.2, 128.6, 128.1, 127.5, 127.1, 127.0, 125.6, 114.7, 70.1, 53.1, 48.5, 47.4, 29.7, 28.4, 27.7, 22.3, 21.6, 14.1 ppm; IR (neat): $\tilde{\nu}$ = 3906.7, 3756.1, 3399.8, 2926.0, 2370.1, 1735.2, 1603.8, 1345.0, 1252.9, 1154.2, 1091.8, 757.2, 659.9, 545.6 cm^{-1}; MS (FAB): *m*/*z* (%): 495 (100) $[M+H]^+$; elemental analysis (%) calcd for $C_{27}H_{30}N_2O_5S$: C 65.57, H 6.11, N 5.66; found: C 65.53, H 6.10, N 5.38.

(3*S*,4*S*)-4-Benzyloxy-*N*-[4-hydroxy-1-(toluene-4-sulfonyl)-azepan-3-yl]-benzamide (24c): Yield = 75 %; R_f = 0.3 (ethyl acetate in hexane, 60 %); m.p. 137 °C; $[\alpha]_D^{25}$ = −10.72 (*c* = 0.25 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ = 7.97 (d, *J* = 8.8 Hz, 2H; Ar*H*), 7.70 (d, *J* = 8.8 Hz, 2H; Ar*H*), 7.47–7.28 (m, 7H; Ar*H*), 7.05 (d, *J* = 8.8 Hz, 2H; Ar*H*), 5.14 (s, 2H; CH_2Ph), 4.48 (s, 1H; N*H*), 4.10 (s, 1H; 3-*H*), 3.86–3.67 (m, 3H; 4-*H*, 2-*H*,7-*H*), 2.97–2.89 (m, 2H; 2′-*H*, 7′-*H*), 2.46 (s, 3H; CH_3), 2.16–1.68 ppm (m, 4H; 5-*H*, 6-*H*); ^{13}C NMR (75 MHz, $CDCl_3$, 25 °C, TMS): δ = 168.4, 160.5, 142.8, 135.1, 133.4, 128.7, 128.2, 127.8, 127.3, 126.8, 126.1, 125.8, 124.4, 120.8, 113.5, 68.8, 51.99, 47.2, 46.3, 28.4, 27.2, 21.1, 20.2 ppm; IR (neat): $\tilde{\nu}$ = 3906.7, 3756.1, 3399.8, 2926.0, 2370.1, 1735.2, 1603.8, 1345.0, 1252.9, 1154.2, 1091.8, 757.2, 659.9, 545.6 cm^{-1}; MS (ESI): *m*/*z* (%): 517.3 (100) $[M+Na]^+$, 495.2 (40) $[M+H]^+$; elemental analysis (%) calcd for $C_{27}H_{30}N_2O_5S$: C 65.57, H 6.11, N 5.66; found: C 65.52, H 6.05, N 5.52.

(3*S*,4*R*)-4-Benzyloxy-*N*-[4-hydroxy-1-(toluene-4-sulfonyl)-azepan-3-yl]-benzamide (24d): Yield = 73 %; R_f = 0.31 (ethyl acetate in hexane, 60 %); m.p. 141 °C; $[\alpha]_D^{25}$ = −3.72 (*c* = 0.345 in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ = 7.94 (d, *J* = 8.8 Hz, 2H; Ar*H*), 7.68 (d, *J* = 8.8 Hz, 2H; Ar*H*), 7.48–7.31 (m, 7H; Ar*H*), 7.02 (d, *J* = 8.8 Hz, 2H; Ar*H*), 5.16 (s, 2H; CH_2Ph), 4.46 (s, 1H; N*H*), 4.02 (s, 1H; 3-*H*), 3.88–3.67 (m, 3H; 4-*H*, 2-*H*, 7-*H*), 2.96–2.90 (m, 2H; 2′-*H*, 7′-*H*), 2.43 (s, 3H; C*H*3), 2.05–1.82 ppm (m, 4H; 5-*H*,6-*H*); IR (neat): $\tilde{\nu}$ = 3906.7, 3756.1, 3399.8, 2926.0, 2370.1, 1735.2, 1603.8, 1345.0, 1252.9, 1154.2, 1091.8, 757.2, 659.9, 545.6 cm^{-1}; MS (ESI): *m*/*z* (%): 517.3 (100) $[M+Na]^+$, 495.2 (40) $[M+H]^+$; elemental analysis (%) calcd for $C_{27}H_{30}N_2O_5S$: C 65.57, H 6.11, N 5.66; found: C 65.47, H 6.01, N 5.54.

3,5-Dibenzyloxy-4-[(2-[1,3]dioxan-2-yl-6-methoxymethoxy-phenyl)-hydroxy-methyl]-benzoic acid *tert*-butyl ester (25): *n*-BuLi (4.5 mL, 1.0 M solution in hexane) was added dropwise at −78 °C under nitrogen to a solution of bromo compound **15** (2 g, 4.27 mmol) in dry THF. The solution was stirred for 5 min, and then a solution of aldehyde **14** (1.0 g, 3.97 mmol) in THF was added dropwise. The reaction mixture was stirred at the same temperature for 1 h and then at room temperature for 4 h. The reaction was quenched with sat. NH_4Cl solution. The organic layer was separated, and the aq. layer was extracted with ethyl acetate. The combined organic layer was washed with brine and dried over anhydrous Na_2SO_4. Chromatography over silica gel afforded alcohol **25** (2.1 g, 79 %) as a sticky solid. R_f = 0.3 (ethyl acetate in hexane, 50 %); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ = 7.45 (d, *J* = 8.7 Hz, 1H; Ar*H*), 7.44–7.21 (m, 12H; Ar*H*), 7.01 (d, *J* = 8.7 Hz. 1H; Ar*H*), 6.69 (d, *J* = 8.7 Hz, 1H; Ar*H*), 6.06 (s, 1H), 5.33 (d, *J* = 8.9 Hz, 1H), 5.11 (s, 4H), 4.80 (s, 2H), 4.16–4.11 (m, 2H), 3.87 (t, *J* = 15 Hz, 1H), 3.68 (t, *J* = 15 Hz, 1H), 2.98 (s, 3H; OCH_3), 2.21–2.09 (m, 1H), 1.58 (s, 9H; OC(CH_3)$_3$), 1.33 ppm (d, *J* = 14 Hz, 1H); MS (ESI): *m*/*z* (%): 665 (59) $[M+Na]^+$, 566.9 (100) $[M-CO_2tBu+Na]^+$; elemental analysis (%) calcd for $C_{38}H_{42}O_9$: C 71.01, H 6.59; found: C 70.71, H 6.54.

3,5-Dibenzyloxy-4-(2-[1,3]dioxan-2-yl-6-methoxymethoxy-benzoyl)-benzoic acid *tert*-butyl ester (26): Alcohol **2** (1.5 g) was taken up in dry CH_2Cl_2 (50 mL). MnO_2 (1.5 g) was added to it in one portion, and the reaction mixture was stirred for 6 h at room temperature. The solid was filtered through Celite, and the filtrate was concentrated. The residue was chromatographed over silica gel to furnish the oxidized product **26** (1.4 g) as a sticky solid. Yield = 93.6 %; R_f = 0.4 (ethyl acetate in hexane, 45 %); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): δ = 7.50 (d, *J* = 9.0 Hz, 1H; Ar*H*), 7.39 (t, *J* = 8.1 Hz, 1H; Ar*H*), 7.30–7.15 (m, 12H; Ar*H*), 7.00 (d, *J* = 9.0 Hz, 1H; Ar*H*), 5.64 (s, 1H), 5.06 (s, 4H), 4.70 (s, 2H), 4.16–4.11 (m, 2H), 3.80 (t, *J* = 9.0 Hz, 2H), 2.94 (s, 3H; OCH_3), 2.21–2.17 (m, 1H), 1.58 (s, 9H; OC(CH_3)$_3$), 1.33 ppm (d, *J* = 14 Hz, 1H); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): δ = 165.1, 157.5, 154.2, 139.2, 136.7, 133.8, 131.2, 128.8, 128.1, 128.5, 121.3, 115.6, 106.7, 99.9, 94.6, 83.1, 70.8, 67.7, 56.4, 28.5 ppm; IR (neat): $\tilde{\nu}$ = 3954, 3906, 3754, 3428, 2820, 2370, 1595, 1351, 1111 cm^{-1}; MS (ESI): *m*/*z* (%): 663.2 (25) $[M+Na]^+$, 641.1 (100) $[M+H]^+$; elemental analysis (%) calcd for $C_{38}H_{40}O_9$: C 71.23, H 6.29; found: C 71.03, H 6.12.

3,5-Dibenzyloxy-4-(2-formyl-6-methoxymethoxybenzoyl)-benzoic acid *tert*-butyl ester (27): Compound **26** (1.2 g, 1.87 mmol) was taken up in acetone/water (25 mL, 9:1 *v*/*v*). A catalytic amount of PTSA was added to it, and the reaction mixture was heated at reflux for 30 min. The solvent was evaporated and the residue was diluted with ethyl acetate. The organic layer was washed with water, followed by brine, and dried over anhydrous Na_2SO_4. Chromatography over silica gel furnished **27** (900 mg) as a white solid. Yield = 86%; R_f = 0.5 (ethyl acetate in hexane, 45%); m.p.: 65°C. ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): δ = 9.90 (s, 1H; C*H*O), 7.56–7.10 (m, 15H; Ar*H*), 5.08 (s, 4H), 4.79 (s, 2H), 3.01 (s, 3H; OCH_3), 1.60 ppm (s, 9H; OC(CH_3)$_3$); IR (neat): $\tilde{\nu}$ = 3954, 3854, 3777, 3408, 2966, 2368, 1595, 1351, 1161, 1104 cm^{-1}; MS (ESI): *m*/*z* (%): 605.1 (100) [*M*+Na]$^+$, 582.9 (40) [*M*+H]$^+$; elemental analysis (%) calcd for $C_{35}H_{34}O_8$: C 72.15, H 5.88; found: C 71.65, H 5.38.

3,5-Dibenzyloxy-4-(2-carboxy-6-methoxymethoxy-benzoyl)-benzoic acid *tert*-butyl ester (4): A solution of aldehyde **27** (700 mg, 1.2 mmol) and NaH_2PO_4 (50 mg, 0.42 mmol) in acetonitrile/water (16 mL, 6:1 *v*/*v*) was cooled to 0°C. H_2O_2 (0.3 mL, 30% solution in water) was added, followed by sodium chlorite (0.44 g). The mixture was stirred for 1 h and the solvent was removed in vacuo. The residue was dissolved in ethyl acetate, and the organic layer was washed with water, followed by brine, and dried over Na_2SO_4. Chromatography over silica gel furnished acid **4** (650 mg, 86%) as a sticky solid. R_f = 0.4 (methanol in chloroform, 5%); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): δ = 8.31 (s, 1H; Ar*H*), 7.44–7.09 (m, 15H; Ar*H*), 5.07 (dd, J_1 = 6.7, J_2 = 35 Hz, 2H), 4.95 (s, 4H), 3.22 (s, 3H; OCH_3), 1.59 ppm (s, 9H; OC(CH_3)$_3$); ^{13}C NMR (75 MHz, $CDCl_3$, 25°C, TMS): δ = 163.2, 157.1, 134.8, 133.6, 129.6, 127.7, 127.1, 127, 126.8, 126.6, 126.2, 126.1, 117.7, 116.6, 105.4, 103.6, 93.1, 92.6, 80.7, 69.5, 54.6, 26.8, 24.1, 21.7, 12.7 ppm; IR (neat): $\tilde{\nu}$ = 3906, 3858, 3806, 3755, 3652, 3425, 2976, 2368, 1675, 1593, 1354, 111.9 cm^{-1}; MS (FAB): *m*/*z* (%): 621.1 (100) [*M*+Na]$^+$, 598.9 (70) [*M*+H]$^+$, 542.9 (40) [*M*−*t*Bu]$^+$; elemental analysis (%) calcd for $C_{35}H_{34}O_9$: C 70.22, H 5.72; found: C 70.02, H 5.51.

Benzyl ester 28: Solid K_2CO_3 (580 mg, 4.2 mmol) was added to a solution of compound **4** (500 mg, 0.84 mmol) in dry acetone (15 mL), followed by benzyl bromide (157 mg, 0.9 mmol). The reaction mixture was heated at reflux at 70°C for 1 h, the solid was filtered off, and the filtrate was concentrated in vacuo. Chromatography over silica gel furnished compound **28** (540 mg, 94%) as a sticky solid. R_f = 0.6 (ethyl acetate in hexane, 25%); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): δ = 7.39–7.11 (m, 20H; Ar*H*), 5.16 (s, 2H), 4.96 (s, 4H), 4.80 (s, 2H), 3.15 (s, 3H; OCH_3), 1.61 ppm (s, 9H; OC(CH_3)$_3$); ^{13}C NMR (75 MHz, $CDCl_3$, 25°C, TMS): δ = 189.9, 165.4, 163.5, 157.10, 153.3, 134.9, 134.6, 133.8, 132.7, 130.3, 128.6, 126.9, 126.6, 126.4, 126.0, 121.8, 117.1, 105.7, 93.3, 80.4, 69.6, 65.6, 54.5, 30.6, 28.4, 28.0, 26.8, 21.4, 12.7 ppm; IR (neat): $\tilde{\nu}$ = 3956, 3906, 3757, 3425, 2816, 2371, 1595, 1351 cm^{-1}; MS (FAB): *m*/*z* (%): 711.2 (100) [*M*+Na]$^+$, 688.9 (89) [*M*+H]$^+$, 632.9 (20) [*M*−*t*Bu]$^+$; elemental analysis (%) calcd for $C_{42}H_{40}O_9$: C 73.24, H 5.85; found: C 73.06, H 5.62.

Acid 5: A solution of compound **28** (500 mg) in dry quinoline (5 mL) was heated at 195°C under nitrogen for 1 h. The reaction mixture was diluted with ether. The organic layer was washed with HCl (2N), followed by brine, and dried over anhydrous Na_2SO_4. Chromatography over silica gel furnished acid **2** (250 mg, 54%) as a sticky brown solid. R_f = 0.3 (5% methanol in chloroform); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): δ = 12.05 (s, 1H; CO_2H), 7.39–7.08 (m, 20H; Ar*H*), 5.13 (d, J = 5.1 Hz, 1H), 4.95 (s, 4H), 4.78 (d, J = 5.1 Hz, 1H), 4.58 (s, 2H), 3.60 ppm (s, 3H; OCH_3); MS (ESI): *m*/*z* (%): 655.1 (100) [*M*+Na]$^+$, 632.8 (39) [*M*]$^+$, 599.0 (9) [*M*−OCH$_3$]$^+$, 576.8 (10) [*M*−*t*Bu]$^+$; elemental analysis (%) calcd for $C_{38}H_{32}O_9$: C 72.14, H 5.10; found: C 72.02, H 4.98.

General Procedure for Mukaiyama esterification: Amido alcohol (1 mmol), acid **5** (1 mmol), and 2-chloro-1-methylpyridinium iodide (1.3 mmol) were taken up in dry CH_2Cl_2 (10 mL). Et_3N (1.5 mmol) was added, and the reaction mixture was stirred at room temperature. A catalytic amount of DMAP was added after 30 min, and the mixture was stirred for 1 h. The solvent was removed, and the residue was chromatographed over silica gel to furnish the coupled product.

(3*R*,4*R*)-3-(4-Benzyloxy-benzoylamino)-4-[3,5-dibenzyloxy-4-(2-benzyloxycarbonyl-6-methoxymethoxy-benzoyl)-benzoyloxy]-azepane-1-carboxylic acid benzyl ester (30a): Yield = 82%; R_f = 0.4 (ethyl acetate in hexane, 45%); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): (3:1 rotameric mixture) δ = 7.64 (d, J = 8.7 Hz, 2H; Ar*H*), 7.49–6.81 (m, 30H; Ar*H*), 6.82 (d, J = 8.7 Hz, 2H; Ar*H*), 5.31 (d, J = 7.9 Hz, 1H), 5.19–4.69 (m, 12H), 3.98 (pseudo t, 1H), 3.71–3.21 (m, 3H), 3.18–3.03 (m, 2H), 3.02 (s, 3H; OCH_3), 2.23–1.64 ppm (m, 4H); IR (neat): $\tilde{\nu}$ = 3864, 3447, 2922, 2358, 1638, 1461, 1380, 1256, 764, 670 cm^{-1}; MS (ESI): *m*/*z* (%): 1091.2 (20) [*M*+Na]$^+$, 1089.2 (100) [*M*]$^+$, 1089.2 (100) [*M*]$^+$; elemental analysis (%) calcd for $C_{66}H_{60}N_2O_{13}$: C 72.78, H 5.55, N 2.57; found: C 72.71, H 5.52, N 2.49.

(3*R*,4*S*)-3-(4-Benzyloxybenzoylamino)-4-[3,5-dibenzyloxy-4-(2-benzyloxycarbonyl-6-methoxymethoxy-benzoyl)-benzoyloxy]-azepane-1-carboxylic acid benzyl ester (30b): Yield = 86%; R_f = 0.4 (ethyl acetate in hexane, 45%); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): (3:1 rotameric mixture)[3] δ = 8.18 (m, 1H), 7.64 (d, J = 8.7 Hz, 2H; Ar*H*), 7.46–7.07 (m, 32H; Ar*H*), 6.82 (d, J = 8.7 Hz, 2H; Ar*H*), 5.31 (d, J = 7.9 Hz, 1H), 5.20–4.76 (m, 12H), 4.18 (br s, 1H), 3.81–3.34 (m, 3H), 3.18–2.98 (m, 2H), 3.08 (s, 3H; OCH_3), 2.16–1.60 ppm (m, 4H); MS (FAB): *m*/*z* (%): 1090.2 (45) [*M*+H]$^+$, 1089.2 (100) [*M*]$^+$; elemental analysis (%) calcd for $C_{66}H_{60}N_2O_{13}$: C 72.78, H 5.55, N 2.57; found: C 72.69, H 5.48, N 2.52.

(3*S*,4*S*)-3-(4-Benzyloxybenzoylamino)-4-[3,5-dibenzyloxy-4-(2-benzyloxycarbonyl-6-methoxymethoxy-benzoyl)-benzoyloxy]-azepane-1-carboxylic acid benzyl ester (30c): Yield = 82%; R_f = 0.41 (ethyl acetate in hexane, 45%); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): (3:1 rotameric mixture) δ = 7.63 (d, J = 8.7 Hz, 2H; Ar*H*), 7.58–6.92 (m, 30H; Ar*H*), 6.85–6.77 (m, 2H; Ar*H*), 5.31 (d, J = 7.9 Hz, 1H), 5.19–4.61 (m, 12H), 4.00 (pseudo t, 1H), 3.86–3.21 (m, 3H), 3.18–3.03 (m, 2H), 3.02 (s, 3H; OCH_3), 2.34–1.64 ppm (m, 4H); MS (FAB): *m*/*z* (%): 1090.1 (70) [*M*+H]$^+$, 1089.2 (100) [*M*]$^+$; elemental analysis (%) calcd for $C_{66}H_{60}N_2O_{13}$: C 72.78, H 5.55, N 2.57; found: C 72.73, H 5.51, N 2.51.

(3*S*,4*R*)-3-(4-Benzyloxybenzoylamino)-4-[3,5-dibenzyloxy-4-(2-benzyloxycarbonyl-6-methoxymethoxy-benzoyl)-benzoyloxy]-azepane-1-carboxylic acid benzyl ester (30d): Yield = 84%; R_f = 0.4 (ethyl acetate in hexane, 45%); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): (3:2 rotameric mixture) δ = 7.64 (d, J = 8.7 Hz, 2H; Ar*H*), 7.39–6.07 (m, 30H; Ar*H*), 6.89 (br s, 2H; Ar*H*), 5.38 (d, J = 7.9 Hz, 1H), 5.16–4.74 (m, 12H), 4.02 (pseudo t, 1H), 3.81–3.41 (m, 3H), 3.21–3.03 (m, 2H), 3.02 (s, 3H; OCH_3), 2.15–1.63 ppm (m, 4H; 5,6-H); MS (FAB): *m*/*z* (%): 1090.2 (65) [*M*+H]$^+$, 1089.2 (100) [*M*]$^+$; elemental analysis (%) calcd for $C_{66}H_{60}N_2O_{13}$: C 72.78, H 5.55, N 2.57; found: C 72.74, H 5.49, N 2.47.

(3*R*,4*R*) derivative (31a): Yield = 83%; R_f = 0.30 (ethyl acetate in hexane, 40%); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): (mix of rotamers 3:2) δ = 7.83 (d, J = 8.8 Hz, 2H; Ar*H*), 7.69 (d, J = 8.8 Hz, 2H; Ar*H*), 7.46–7.04 (m, 27H; Ar*H*), 6.91 (d, J = 8.8 Hz, 2H; Ar*H*), 5.32 (d, J = 8.3 Hz, 1H), 5.10 (s, 2H; CH_2OCH$_3$), 4.99 (s, 2H; CH_2Ph), 4.96 (s, 2H; CH_2Ph), 4.81–4.80 (m, 1H), 4.76 (s, 2H; OCH_2Ph), 3.98–3.91 (m, 1H), 3.73 (dd, J_1 = 3.99 Hz, J_2 = 14.7 Hz, 1H), 3.15 (dd, J_1 = 3.3 Hz, J_2 = 14.7 Hz, 1H), 3.05 (s, 3H; CH$_2$OCH_3), 2.97–2.91 (m, 1H), 2.43 (s, 3H; CH_3), 2.32–2.29 (m, 1H), 2.22–2.04 (m, 2H), 1.98–1.83 ppm (m, 2H); IR (neat): $\tilde{\nu}$ = 3931.0, 3906.0, 3857.8, 3756.1, 3425.7, 2371.3, 1597.3, 1350.2, 1158.0, 1110.3, 1016.0, 753.5, 695.8 cm^{-1}; MS (ESI): *m*/*z* (%): 1090.2 (60) [*M*+H]$^+$, 1089.2 (100) [*M*]$^+$; elemental analysis (%) calcd for $C_{65}H_{60}N_2O_{13}S$: C 70.38, H 5.45, N 2.53; found: C 70.32, H 5.39, N 2.48.

(3*R*,4*S*) derivative 31b: Yield = 89%; R_f = 0.32 (ethyl acetate in hexane, 40%); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): (mix of rotamers) δ = 7.81 (d, J = 8.8 Hz, 2H; Ar*H*), 7.67 (d, J = 8.25 Hz, 2H; Ar*H*), 7.38–7.02 (m, 27H; Ar*H*), 6.89 (d, J = 8.8 Hz, 2H; Ar*H*), 5.29 (br s, 1H), 5.08 (s, 2H; CH_2OCH$_3$), 4.97 (s, 2H; CH_2Ph), 4.90 (s, 2H; CH_2Ph), 4.81–4.78 (m, 1H), 4.74 (s, 2H; OCH_2Ph), 3.97–3.90 (m, 1H), 3.71 (dd, J_1 = 4.18 Hz, J_2 = 14.6 Hz, 1H), 3.14 (dd, J_1 = 3.14 Hz, J_2 = 14.7 Hz, 1H), 3.06 (s, 3H; CH$_2$OCH_3), 2.97–2.90 (m, 1H), 2.43 (s, 3H; CH_3), 2.34–2.25 (m, 1H), 2.22–2.09 (m, 2H), 1.98–1.87 ppm (m, 2H); IR (neat): $\tilde{\nu}$ = 3931.0, 3906.0, 3857.8, 3756.1, 3425.7, 2371.3, 1597.3, 1350.2, 1158.0, 1110.3, 1016.0, 753.5, 695.8 cm^{-1}; MS (ESI): *m*/*z* (%): 1090.2 (60) [*M*+H]$^+$, 1089.2 (100) [*M*]$^+$; elemental analysis (%) calcd for $C_{65}H_{60}N_2O_{13}S$: C 70.38, H 5.45, N 2.53; found: C 70.31, H 5.41, N 2.45.

(3S,4S) derivative 31c: Yield = 93%; R_f = 0.31 (ethyl acetate in hexane, 40%); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): (mix of rotamers) δ = 7.81 (d, J = 8.8 Hz, 2H; Ar*H*), 7.66 (d, J = 8.25 Hz, 2H; Ar*H*), 7.38–7.02

(m, 27H; Ar*H*), 6.89 (d, $J=8.8$ Hz, 2H; Ar*H*), 5.31 (brs, 1H), 5.10 (s, 2H; CH_2OCH$_3$), 4.98 (s, 2H; CH_2Ph), 4.90 (s, 2H; CH_2Ph), 4.81–4.78 (m, 1H), 4.76 (s, 2H; OCH_2Ph), 3.98–3.89 (m, 1H), 3.72 (dd, $J_1=4.17$ Hz, $J_2=14.8$ Hz, 1H), 3.14 (dd, $J_1=3.36$ Hz, $J_2=14.5$ Hz, 1H), 3.06 (s, 3H; CH$_2$OCH_3), 2.97–2.90 (m, 1H), 2.43 (s, 3H; CH_3), 2.34–2.25 (m, 1H), 2.22–2.09 (m, 2H), 1.98–1.87 ppm (m, 2H); IR (neat): $\tilde{\nu}=3931.0$, 3906.0, 3857.8, 3756.1, 3425.7, 2371.3, 1597.3, 1350.2, 1158.0, 1110.3, 1016.0, 753.5, 695.8 cm^{-1}; MS (ESI): *m*/*z* (%): 1109.0 (100) [*M*+Na]$^+$; elemental analysis (%) calcd for $C_{65}H_{60}N_2O_{13}S$: C 70.38, H 5.45, N 2.53; found: C 70.30, H 5.31, N 2.51.

(3*S*,4*R*) derivative 31d: Yield=87%; $R_f=0.31$ (ethyl acetate in hexane, 40%); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): (mix of rotamers 3:2) $\delta=7.83$ (d, $J=8.8$ Hz, 2H; Ar*H*), 7.69 (d, $J=8.8$ Hz, 2H; Ar*H*), 7.40–7.09 (m, 27H; Ar*H*), 6.92 (d, $J=8.8$ Hz, 2H; Ar*H*), 5.31 (d, $J=8.3$ Hz, 1H), 5.09 (s, 2H; CH_2OCH$_3$), 4.98 (s, 2H; CH_2Ph), 4.91 (s, 2H; CH_2Ph), 4.81–4.80 (m, 1H), 4.76 (s, 2H; OCH_2Ph), 3.98–3.92 (m, 1H), 3.73 (dd, $J_1=3.99$ Hz, $J_2=14.7$ Hz, 1H), 3.15 (dd, $J_1=3.3$ Hz, $J_2=14.7$ Hz, 1H), 3.07 (s, 3H; CH$_2$OCH_3), 2.97–2.91 (m, 1H), 2.45 (s, 3H; CH_3), 2.32–2.29 (m, 1H), 2.22–2.04 (m, 2H), 1.98–1.83 ppm (m, 2H); IR (neat): $\tilde{\nu}=$ 3931.0, 3906.0, 3857.8, 3756.1, 3425.7, 2371.3, 1597.3, 1350.2, 1158.0, 1110.3, 1016.0, 753.5, 695.8 cm^{-1}; MS (ESI): *m*/*z* (%): 1090.2 (60) [*M*+H]$^+$, 1089.2 (100) [*M*]$^+$; elemental analysis (%) calcd for $C_{65}H_{60}N_2O_{13}S$: C 70.38, H 5.45, N 2.53; found: C 70.29, H 5.35, N 2.46.

General procedure for deprotection: A catalytic amount of HCl was added to a solution of a fully protected balanol derivative in methanol. The reaction mixture was heated to 50°C and stirred at same temperature for 3 h. The solvent was evaporated, and the residue was debenzylated by Nicolaou's procedure[39] to afford the balanol derivative.

(3*R*,4*R*)-Balanol (1a): $[\alpha]_D^{25}=-107$ ($c=0.252$ in methanol); ^{1}H NMR (300 MHz, CD_3OD, 25°C, TMS): $\delta=7.60$ (d, $J=8.7$ Hz, 2H; Ar*H*), 7.25 (d, $J=7.8$ Hz, 1H; Ar*H*), 7.17 (t, $J=7.8$ Hz, 1H; Ar*H*), 6.92 (s, 2H; Ar*H*), 6.80 (d, $J=7.8$ Hz, 1H; Ar*H*), 6.76 (d, $J=8.7$ Hz, 2H; Ar*H*), 5.29 (m, 1H; 4-H), 4.32 (brm, 1H; 3-H), 3.42–2.98 (brm, 4H; 2,7-H), 1.84–2.12 ppm (brm, 4H; 5,6-H); MS (ESI): *m*/*z* (%): 551.6 (100) [*M*+H]$^+$; elemental analysis (%) calcd for $C_{28}H_{26}N_2O_{10}$: C 61.09, H 4.76, N 5.09; found: C 60.99, H 4.72, N 5.01.

(3*R*,4*S*)-Balanol (1b): $[\alpha]_D^{25}=-64$ ($c=0.252$ in methanol); ^{1}H NMR (300 MHz, CD_3OD, 25°C, TMS): $\delta=7.60$ (d, $J=8.7$ Hz, 2H; Ar*H*), 7.25 (d, $J=7.8$ Hz, 1H; Ar*H*), 7.17 (t, $J=7.8$ Hz, 1H; Ar*H*), 6.92 (s, 2H), 6.80 (d, $J=7.8$ Hz, 1H; Ar*H*), 6.76 (d, $J=8.7$ Hz, 2H; Ar*H*), 5.30 (m, 1H; 4-H), 4.45 (brm, 1H; 3-H), 3.32–2.97 (brm, 4H; 2,7-H), 2.12–1.78 ppm (brm, 4H; 5,6-H); MS (ESI): *m*/*z* (%): 551.2 (100) [*M*+H]$^+$; elemental analysis (%) calcd for $C_{28}H_{26}N_2O_{10}$: C 61.09, H 4.76, N 5.09; found: C 60.89, H 4.69, N 5.03.

(3*S*,4*S*)-Balanol (1c): $[\alpha]_D^{25}=+97.8$ ($c=0.331$ in methanol); ^{1}H NMR (300 MHz, CD_3OD, 25°C, TMS): $\delta=7.60$ (d, $J=8.7$ Hz, 2H; Ar*H*), 7.25 (d, $J=7.8$ Hz, 1H; Ar*H*), 7.17 (t, $J=7.8$ Hz, 1H; Ar*H*), 6.92 (s, 2H), 6.80 (d, $J=7.8$ Hz, 1H; Ar*H*), 6.76 (d, $J=8.7$ Hz, 2H; Ar*H*), 5.19 (m, 1H; 4-H), 4.35 (brm, 1H; 3-H), 3.47–3.03 (brm, 4H; 2,7-H), 2.12–1.74 (brm, 4H; 5,6-H); MS (ESI): *m*/*z* (%): 551.5 (100) [*M*+H]$^+$; elemental analysis (%) calcd for $C_{28}H_{26}N_2O_{10}$: C 61.09, H 4.76, N 5.09; found: C 60.91, H 4.67, N 5.00.

(3*S*,4*R*)-Balanol (1d): $[\alpha]_D^{25}=+54$ ($c=0.321$ in methanol); ^{1}H NMR (300 MHz, CD_3OD, 25°C, TMS): $\delta=7.60$ (d, $J=8.7$ Hz, 2H; Ar*H*), 7.25 (d, $J=7.8$ Hz, 1H; Ar*H*), 7.17 (t, $J=7.8$ Hz, 1H; Ar*H*), 6.92 (s, 2H), 6.80 (d, $J=7.8$ Hz, 1H; Ar*H*), 6.76 (d, $J=8.7$ Hz, 2H; Ar*H*), 5.51 (m, 1H; 4-H), 4.51 (brm, 1H; 3-H), 3.32–3.07 (brm, 4H; 2,7-H), 2.12–1.78 ppm (brm, 4H; 5,6-H); MS (ESI): *m*/*z* (%): 551.6 (100) [*M*+H]$^+$; elemental analysis (%) calcd for $C_{28}H_{26}N_2O_{10}$: C 61.09, H 4.76, N 5.09; found: C 60.94, H 4.70, N 4.91.

***N*-Tosyl-(*R*,*R*)-balanol (2a)**: Yield=89%; $R_f=0.3$ (MeOH in $CHCl_3$, 60%); $[\alpha]_D^{25}=-48.3$ ($c=0.121$ in methanol); ^{1}H NMR (300 MHz, CD_3OD, 25°C, TMS): $\delta=7.76$ (t, $J=7.9$ Hz, 3H; Ar*H*), 7.66 (d, $J=$ 8.6 Hz, 1H; Ar*H*), 7.43 (t, $J=7.32$ Hz, 2H; Ar*H*), 7.22 (t, $J=8.5$ Hz, 2H; Ar*H*), 6.94 (d, $J=7.8$ Hz, 1H; Ar*H*), 6.93 (s, 2H; Ar*H*), 6.80 (d, $J=$ 8.7 Hz, 2H; Ar*H*), 5.38 (d, $J=9.3$ Hz, 1H; 4-H), 4.51 (brm, 1H; 3-H), 3.56 (brs, 2H; 2,7-H), 3.42 (brs, 2H; 2,7-H), 2.44 (s, 3H; CH_3), 2.37–2.21 (brm, 2H; 5,6-H), 1.93–1.87 ppm (brm, 2H; 5,6-H); IR (neat): $\tilde{\nu}=$ 3827.8, 3751.1, 3421.7, 2371.3, 1697.3, 1351.2, 1058.0, 1016.0, 753.5, 695.8 cm^{-1}; MS (ESI): *m*/*z* (%): 727.2 (100) [*M*+Na]$^+$; elemental analysis (%) calcd for $C_{35}H_{32}N_2O_{12}S$: C 59.65, H 4.58, N 3.98; found: C 59.59, H 4.51, N 3.78.

(3*R*,4*R*)-3-*tert*-Butoxycarbonylamino-4-hydroxy-azepane-1-carboxylic acid benzyl ester (32): Compound **21a** (70 mg, 0.172 mmol) was taken up in dry CH_2Cl_2 (10 mL), trimethylsilyl bromide (0.034 mL, 0.258 mmol) was added at 0°C, and the reaction mixture was stirred at room temperature for 4 h. The solvent was removed, and the residue was dissolved in diethyl ether. The organic layer was washed with aqueous $NaHCO_3$, followed by brine, and the organic layer was dried on anh. Na_2SO_4 and evaporated in vacuo. The residue was passed through a small silica pad and was used for the next step without further purification. Crude yield: 75%; $R_f=0.4$ (ethyl acetate in hexane, 35%), Crude MS (ESI): *m*/*z* (%): 387.2 (100) [*M*+Na]$^+$; elemental analysis (%) calcd for $C_{19}H_{28}N_2O_5$: C 62.62, H 7.74, N 7.69; found: C 62.58, H 7.67, N 7.59.

(3*R*,4*R*)-4-(4-Benzyloxybenzoyloxy)-3-*tert*-butoxycarbonylamino-azepane-1-carboxylic acid benzyl ester (33): Compound **32** (50 mg, 0.137 mmol), *p*-benzyloxy-benzoic acid (36.2 mg, 0.164 mmol), and 2-chloro-1-methylpyridinium iodide (52.5 mg, 0.206 mmol) were taken up in dry CH_2Cl_2 (5 mL). Triethylamine (0.04 mL, 0.274 mmol) was added, and the reaction mixture was stirred for one hour. After completion of the reaction, solvent was evaporated, and the residue was chromatographed to furnish the pure product **33** as a sticky solid. Yield=75 mg (95%); $R_f=0.6$ (ethyl acetate in hexane, 40%); ^{1}H NMR (300 MHz, $CDCl_3$, 25°C, TMS): $\delta=7.97$ (d, $J=8.7$ Hz, 2H; Ar*H*), 7.93–7.31 (m, 10H; Ar*H*), 6.97 (d, $J=8.7$ Hz, 2H; Ar*H*), 5.28–5.08 (m, 4H), 4.16 (brs, 1H), 3.68–3.50 (brm, 4H), 2.18 (brs, 1H), 1.98–1.61 (brm, 4H), 1.40 ppm (s, 9H; -OC(CH_3)$_3$); ^{13}C NMR (75 MHz, $CDCl_3$, 25°C, TMS): $\delta=164.3$, 153.7, 138.5, 130.4, 127.4, 127.2, 126.9, 126.6, 126.4, 126.1, 113.3, 78.4, 68.8, 66.3, 66.1, 45.8, 28.4, 27.0 ppm; IR (neat): $\tilde{\nu}=3766$, 3452, 3221, 1736, 1692, 1112, 760 cm^{-1}; MS (ESI): *m*/*z* (%): 597.1 (100) [*M*+Na]$^+$, 575.2 (75) [*M*+H]$^+$, 418 (60) [*M*−*t*Bu]$^+$, 475.2 (45) [*M*−*t*Boc]$^+$; elemental analysis (%) calcd for $C_{33}H_{38}N_2O_7$: C 68.97, H 6.67, N 4.87; found: C 68.91, H 6.59, N 4.74.

Fully protected ophiocordin (35): Compound **33** (25 mg, 0.05 mmol) was taken up in dry CH_2Cl_2. TFA in CH_2Cl_2 (5%) was added, and the reaction mixture was stirred at room temperature for 2 h. After consumption of all of the starting material, solvent was evaporated. Co-evaporation was performed twice with CH_2Cl_2 to remove the excess TFA. The residue was dried and again dissolved in CH_2Cl_2. Acid **4** (30 mg, 0.05 mmol) and 2-chloro-1-methylpyridinium iodide (27 mg, 0.105 mmol) were added, followed by tri-*n*-butylamine (0.035 mL, 0.150 mmol), and the reaction mixture was stirred at room temperature. A catalytic amount of DMAP was added after 30 min, and the system was stirred for 1 h. The solvent was removed, and the residue was chromatographed over silica gel to furnish the coupled product **35** (7 mg, 35%). Some starting material **34** was recovered (15 mg). (3:1.5 rotameric mixture); ^{1}H NMR (600 MHz, $CDCl_3$, 25°C, TMS): $\delta=8.02$ (d, $J=12$ Hz, 2H; Ar*H*), 7.49–7.00 (m, 43H; Ar*H*), 5.16–4.92 (m, 10H), 4.22 (brs, 2H), 3.78–3.6 (m, 4H), 3.31 (s, 3H), 3.29 (s, 3H), 2.43–2.31 (m, 1H), 2.21–1.8 (m, 4H), 1.55 ppm (s, 9H); MS (ESI): *m*/*z* (%): 1077.2 (10) [*M*+Na]$^+$, 1055.1 (100) [*M*+H]$^+$, 999.1 (15) [*M*−*t*Bu]$^+$; elemental analysis (%) calcd for $C_{63}H_{62}N_2O_{13}$: C 71.71, H 5.92, N 2.65; found: C 71.65, H 5.87, N 2.60.

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Original article

Thiophene containing trisubstituted methanes [TRSMs] as identified lead against *Mycobacterium tuberculosis*

Priyanka Singh [a], Sudipta Kumar Manna [a], Amit Kumar Jana [a], Tiash Saha [a], Pankaj Mishra [a], Saurav Bera [a], Maloy Kumar Parai [a], Srinivas Lavanya Kumar M [a], Sankalan Mondal [a], Priyanka Trivedi [b], Vinita Chaturvedi [b], Shyam Singh [b], Sudhir Sinha [b, *], Gautam Panda [a, *]

[a] Medicinal and Process Chemistry Division, CSIR-Central Drug Research Institute, B.S. 10/1, Jankipuram Extension, Sitapur Road, Lucknow-226031, UP, India
[b] Biochemistry Division, CSIR-Central Drug Research Institute, B.S. 10/1, Jankipuram Extension, Sitapur Road, Lucknow-226031, UP, India

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ABSTRACT

Triarylmethanes (TRAMs) and thiophene containing trisubstituted methanes (TRSMs) have been reported by us, having potential against *Mycobacterium tuberculosis* and *Mycobacterium fortuitum* strains, respectively. Further, extension through synthesis and biological evaluation of novel TRSMs resulted into an identified lead 36 (S006-830) [(diisopropyl-(2-{4-[(4-methoxy-phenyl)- thiophen-2-yl-methyl]-phenoxy}-ethyl)-amine)] with MIC: 1.33 mg/L, non-toxic against Vero C-1008 cell line with selectivity index >10, *ex vivo* efficacy equivalent to first line TB drugs-isoniazid (INH), rifampicin (RFM) and pyrazinamide (PZA) in the mouse and human macrophages, and lung CFU count of 2.2×10^7 (approximately 15 fold lesser than untreated mice, 31×10^7) with efficacies comparable to ethambutol (EMB) (1.27×10^7) and PZA (1.9×10^7). Further, S006-830 also showed potent bactericidal activity against multi-drug resistant and single-drug resistant clinical isolates of *M. tuberculosis*.

1. Introduction

Tuberculosis (TB), caused by *Mycobacterium tuberculosis* (*M. tb*) is not merely a disease rather it is an epidemic which is prevalent in modern era of mankind and is considered as the world's deadliest communicable disease by World Health Organization (WHO) [1,2]. Worldwide, according to WHO report 2014, about 9 million people were infected with TB in the year 2013 and an approximate of 1.4 million died in a single year of 2011 [1,2] and the scenario is being worsen by multidrug-resistant (MDR) and extensively drug-resistant (XDR) TB, often emerged from the patient's poor compliance to the standard drugs therapy regimen. Hence, discovery of newer inhibitors of *M. tb* is a global and urgent need. In our on-going campaign against TB, we previously reported triarylmethanes (TRAMs) [3–6] and thiophene containing trisubstitued methanes (TRSMs) [7] as novel cores having anti-tubercular activity (Fig. 1). In 2012, we reported the potential of few novel TRSMs against acute and persistent infection of *Mycobacterium fortuitum* (responsible for the opportunist non-tubercular infection in humans) in a murine infection model and identified two promising lead compounds (Fig. 1) [8]. Further, optimization through synthetic and medicinal chemistry approach led us to present the current work with potential applicability of novel TRSMs as antimycobacterial compounds.

2. Results and discussion

2.1. Chemistry

The reaction scheme followed the similar protocol as outlined in our previous report [7,8]. We emphasized on the design, synthesis and anti-tubercular activity of thiophene containing TRSMs with alkylaminoethyl chains **A**.

In this work, we have emphasized on the design, synthesis and anti-tubercular activity of thiophene containing TRSMs with alkylaminoethyl chains A (Fig. 2). Initially, the Grignard reagent aryl magnesium bromide **1a**–**j** were reacted with various commercially available thiophene-2-aldehyde **2** to synthesize carbinol **3**. Then

* Corresponding authors.
E-mail addresses: sinha.sudhir@gmail.com, sudhir_sinha@cdri.res.in (S. Sinha), gautam.panda@gmail.com, gautam_panda@cdri.res.in (G. Panda).

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0223-5234/

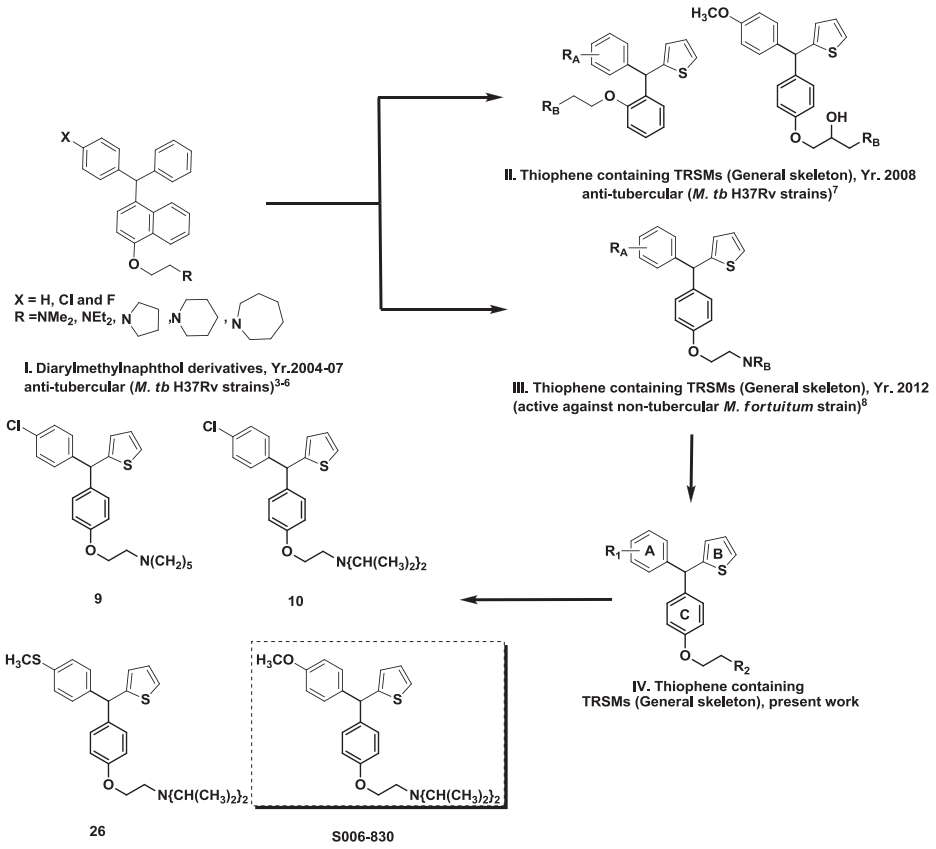


Fig. 1. Evolution of triarylmethanes into thiophene containing TRSMs identified potential lead **36** (S006-830) against *M. tuberculosis*.

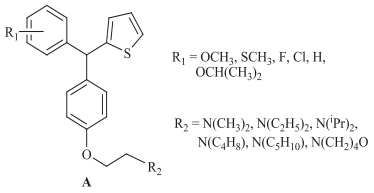


Fig. 2. General skeleton of designed molecules.

Friedel–Crafts alkylation of **3** with phenol in the presence of conc. H_2SO_4 in dry benzene furnished **4** in satisfactory yields. Alkylated product **4** was refluxed with various dialkylaminoethyl chloride hydrochloride chains in presence of the K_2CO_3 in acetone to obtain compounds **5**–**36** in good yields, (Schemes 1 and 2).

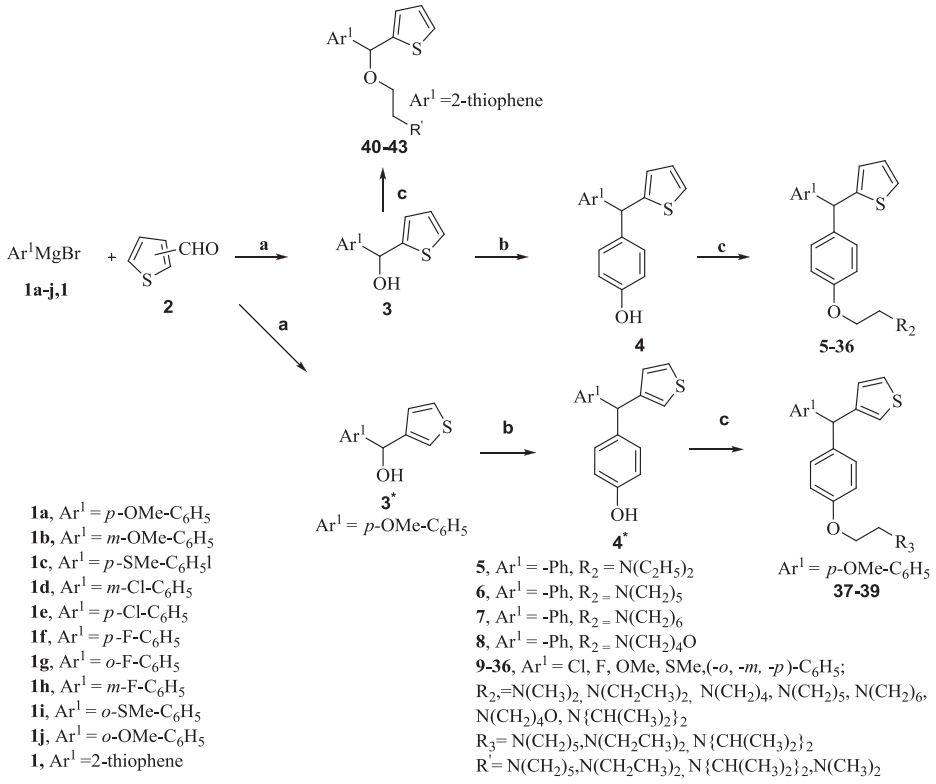
Later to analyze the activity of dithiophene compounds several compounds with 2-thiophene substitution were synthesized by taking commercially available 2-thiophene carboxaldehyde and 2-substituted thiophene bromide with magnesiun in THF to prepare respective carbinol followed by refluxing it with various dialkylaminoethyl chloride hydrochloride chains with K_2CO_3 as base in acetone to obtain the final compounds **40**–**43**. Similarly, 3-thiophene substituted compounds were prepared from 3-thiophene carboxaldehyde and 3-thiophene bromide by the similar way as above to obtain final compounds **44**–**49**.

Initially, synthesis of compounds without any substitution on benzene ring were carried out by nucleophilic addition of phenylmagnesium bromide on 2-thiophene carbaldehyde to provide phenyl thiophen-2-yl carbinol. After this similar protocol was followed as above to synthesize compounds **5**–**8** (Table 1). To explore the effect of substitution on benzene ring, various substitutions were incorporated in the basic framework like halogen, methoxy and thiomethoxy substituted arylbromide to synthesize the grignard reagent and then compounds **9**–**36** (Table 1) were synthesized by procedure stated above. Similarly, to understand the effect of substitution at 3 position of thiophene, 3-thiophene carboxaldehyde was also incorporated to provide *p*-methoxy substituted phenyl thiophen-3-yl carbinol which were used subsequently to synthesize compounds **37**–**39** by above mentioned procedure. Later dithiophene substituted compounds **40**–**49** were also synthesized by the same procedure.

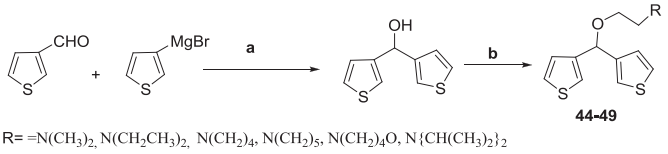
2.2. Biological evaluation

2.2.1. In vitro screening of compounds using virulent strain of *M. tuberculosis* to select 'hits'

This assay measures the capability of the test compound to kill or inhibit the multiplication of pathogenic *M. tuberculosis* H37Rv and is considered as 'primary screening method'. Compounds showing MIC ≤6.25 mg/L were considered for further evaluation [11]. From our previous report [7], it was evident that the amino alkyl chain (Fig. 1II) exhibited the maximum impact on the anti-tubercular activity (MIC <12.5 mg/L) and *para*-substituted chain (R_B) over the ring **C** remains critical for the activity as the *ortho*-substituted derivatives demonstrated very poor activity (MIC: *para*-substituted = < 12.5 mg/L; *ortho*-substituted = >12.5 mg/L). Surprisingly, both electron-donating group (EDG) and electron withdrawing group (EWG) substituted over the *para*/*meta*-position (R_B) of ring A exhibited good anti-tubercular activity. In line with the observations from the previous report, we decided to continue our



Scheme 1. Synthesis of Trisubstituted Methanes (TRSMs). Reagents and conditions (a) Dry THF, 0 °C-rt, 30 min; (b) Phenol, dry benzene, catalytic H_2SO_4, reflux, 1 h; (c) Alkylaminoethyl hydrochloride, dry acetone, anhyd. K_2CO_3, reflux, 6–7 h.



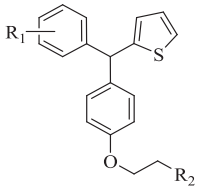
Scheme 2. Synthesis of dithiophene substituted compounds. Reagents and conditions (a) Dry THF, 0 °C-rt, 30 min; 1 h; (b) Alkylaminoethyl hydrochloride, dry acetone, anhyd. K_2CO_3, reflux, 6–7 h.

work by changing the best possible substitutions on the amino alkyl chain R_2 on ring C and observed their anti-tubercular effects. All thirty one compounds were found to be active against *M. tuberculosis* H37Rv with MIC ranging from 0.78 to 25.0 mg/L (Table 1). Nine compounds [MIC: **9** = 0.78 mg/L, **15** = 1.34 mg/L, **12** = 1.25 mg/L, **20** = 1.24 mg/L, **32** = 1.28 mg/L, **14** = 1.29 mg/L, **10** = 1.56 mg/L, **26** = 1.57 mg/L, and **S006-830** = 1.33 mg/L] demonstrated notable inhibitory capacity against *M. tuberculosis* $H37R_v$ strain. While ring A substituted with *para*-chloro and $N(CH_2)_5$ as amino alkyl chain **9** showed the most potent inhibition with MIC of 0.78 mg/L, *meta*-substituted counterpart **14** showed somewhat diminished activity MIC = 1.29 mg/L. While *ortho*-fluoro substituted compound **20** with the similar alkyl chain [$N(CH_2)_5$] exhibited diminished inhibition [MIC = 1.24 mg/L] as compared to the chloro substituted derivative **9**, the *meta*-fluoro substituted compound **25** showed highly diminished activity [MIC = 19.8 mg/L]. Towards the observation with EDGs at ring A, somewhat comparable inhibition has been observed with compound **32** carrying *ortho*-OCH_3 with [$N(CH_2)_5$] side chain with MIC = 1.28 mg/L. Additionally, compound **36 (S006-830)** with *para*-OCH_3 and N{$CH(CH_3)_2$}$_2$ demonstrated potent inhibition of *M. tuberculosis* $H37R_v$ strain with MIC = 1.33 mg/L as compared to their *ortho*-OCH_3 substituted counterpart **34** with very low activity (25.0 mg/L). Surprisingly, all 3-substituted compounds (**37**–**39**, **44**–**49**) exhibited low activity (25.0 mg/L). Moreover, 2-substituted dithiophene containing compounds also exhibited moderate activity (12.5 mg/L).

2.2.2. *Cytotoxicity assessment through in vitro toxicity assay and ex vivo activity in the mouse and human macrophages model of tuberculosis*

Initially, *in vitro* cytotoxicity of active compounds (having MIC ≤ 6.25 mg/L) was measured using Vero cells (Vero C-1008 cell line) and if found non-toxic, were then taken further for their testing by the *ex vivo* assay using mouse bone marrow derived (MBMD) or human monocyte derived macrophage models of tuberculosis. The macrophage model mimics growth environment of natural infection and demonstrates the ability of the candidate molecule to penetrate host cell membrane, phagocytic vacuole and bacilli residing within the vacuole, and hence gives the advantage to reach the desired drug targets. In addition, it also serves as a model for hypoxia induced latent TB infection, since tissue concentration of oxygen is considerably lower than that in the ambient air. Many latency marker proteins of the pathogen are known to get expressed during its intracellular regime. From Vero C-1008 cell line studies, four compounds viz. **9**, **10**, **26** and **36** (**S006-830**) could be evidenced to be non-toxic demonstrating SI of greater than 10 and taken further for their assessment in the MBMDmacrophage model. All four compounds (at 4× MIC) killed greater than 90% of the intracellular *M. tuberculosis*, as judged by counting the viable bacilli (colony forming units or CFUs) in lysates of the drug-treated or untreated infected macrophages. As assessed from the statistical data, the *ex vivo* efficacy of **9**, **10**, **26** and **S006-830** were comparable to that of the first line TB drugs-INH, RFM and PZA (Fig. 2). Two

Table 1
Biological Activity of synthesized TRSMs (**5**–**36**).



9-36

Compd. no.	R_1	R_2	Mol. Weight	CLogP	MIC (mg/L)	Cytotoxic activity, IC_{50} (mg/L)	SI[a]
5	H	$N(C_2H_5)_2$	365.53	6.36	12.47	ND	ND[b]
6	H	$N(CH_2)_5$	377.54	6.44	6.23	ND	ND
7	H	$N(CH_2)_6$	391.57	6.97	3.10	13	<10
8	H	$N(CH_2)_4O$	379.52	5.11	24.98	ND	ND
9	***p*-Cl**	**$N(CH_2)_5$**	**411.99**	**7.15**	**0.78**	**100**	**>10**
10	***p*-Cl**	**$N\{CH(CH_3)_2\}_2$**	**428.03**	**7.69**	**1.56**	**100**	**>10**
11	*m*-Cl	$N(CH_3)_2$	371.92	6.02	4.65	10.8	<10
12	*m*-Cl	$N(CH_2CH_3)_2$	399.98	7.08	1.25	10.1	<10
13	*m*-Cl	$N(CH_2)_4$	397.96	6.62	2.49	8.6	<10
14	*m*-Cl	$N(CH_2)_5$	411.99	7.15	1.29	8.7	<10
15	*m*-Cl	$N\{CH(CH_3)_2\}_2$	428.03	7.69	1.34	22.0	>10
16	*m*-Cl	$N(CH_2)_4O$	413.96	5.83	5.18	23.4	<10
17	*o*-F	$N(CH_3)_2$	355.57	5.45	17.8	ND	ND
18	*o*-F	$N(CH_2CH_3)_2$	383.52	6.51	4.80	ND	ND
19	*o*-F	$N(CH_2)_4$	381.51	6.05	19.1	ND	ND
20	*o*-F	$N(CH_2)_5$	395.53	6.58	1.24	3.9	<10
21	*o*-F	$N\{CH(CH_3)_2\}_2$	411.58	7.12	10.3	ND	ND
22	*o*-F	$N(CH_2)_4O$	397.51	5.26	19.9	ND	ND
23	*m*-F	$N(CH_3)_2$	355.47	5.45	17.8	ND	ND
24	*m*-F	$N(CH_2CH_3)_2$	383.52	6.51	2.40	5.8	<10
25	*m*-F	$N(CH_2)_5$	393.52	6.58	19.8	ND	ND
26	***p*-SCH_3**	**$N\{CH(CH_3)_2\}_2$**	**439.68**	**7.18**	**1.57**	**100**	**>10**
27	*o*-SCH_3	$N(CH_3)_2$	383.57	5.16	12.5	ND	ND
28	*o*-SCH_3	$N(CH_2CH_3)_2$	411.62	6.22	25.0	ND	ND
29	*o*-OCH_3	$N(CH_3)_2$	367.50	4.82	9.19	ND	ND
30	*o*-OCH_3	$N(CH_2CH_3)_2$	395.56	5.88	25.0	ND	ND
31	*o*-OCH_3	$N(CH_2)_4$	393.54	5.43	25.0	ND	ND
32	*o*-OCH_3	$N(CH_2)_5$	407.57	5.96	1.28	9.2	<10
33	*o*-OCH_3	$N(CH_2)_6$	421.59	6.49	2.64	12.7	<10
34	*o*-OCH_3	$N\{CH(CH_3)_2\}_2$	423.61	6.50	25.0	ND	ND
35	*o*-OCH_3	$N(CH_2)_4O$	409.54	4.63	10.24	ND	ND
36 (S006-830)	***p*-OCH_3**	**$N\{CH(CH_3)_2\}_2$**	**423.61**	**6.90**	**1.33**	**100**	**>10**

Compounds showing significant activity have been highlighted in bold.
[a] SI = Selectivity Index.
[b] ND = Not Determined.

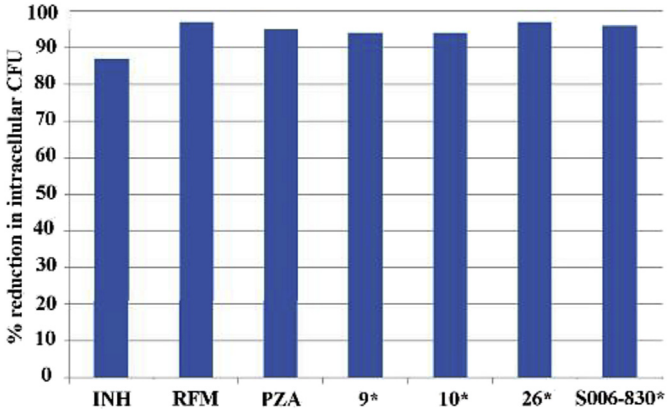


Fig. 3. Percent reduction in intracellular CFU of *M. tuberculosis* $H37R_V$ in mouse bone marrow derived macrophages by **9, 10, 26** and **S006-830**. Isoniazid (INH), rifampicin (RFM) and pyrazinamide (PZA) were used as standard drugs.

compounds **26** and **S006-830** showed *ex vivo* activity comparable to that of the standard drug INH (INH) in human macrophages experimentally infected with *M. tuberculosis* H37Rv (Figs. 3 and 4).

2.2.3. In vivo assay against M. tuberculosis $H37R_V$ infected mouse model

When infected with a high number of *M. tuberculosis* CFUs by the *i.v.* route, the mouse harbours a bacillary population that is similar in number and in metabolic state to that present in the lung cavity of human TB. Thus, the mouse model is able to reproduce bacteriologic conditions close to those present in the natural human disease and provide information on compound activity that can be extrapolated to humans. The most frequently used is the outbred 'Swiss' mouse. The differences in the immune status among outbred Swiss mice also exist among humans, and a drug or regimen that is active against the mycobacteria in Swiss mouse is likely to be active in humans. The strain of choice for tubercle bacilli in the mouse model is the $H37R_v$ strain of *M. tuberculosis*, which maintains virulence through regular passages in the mouse. Efficacy of a compound is monitored by survival/mortality rate, evaluation of body weight, extent of gross lesions, and the enumeration of the CFUs in organs. Usually the CFU counts are performed in the spleen or lungs or both. Since spleen and lung display similar overall results, it is unnecessary to enumerate the CFU in both organs. In general, effective dosages of compounds (in mg/kg body weight) in the laboratory animals are larger than those in humans.

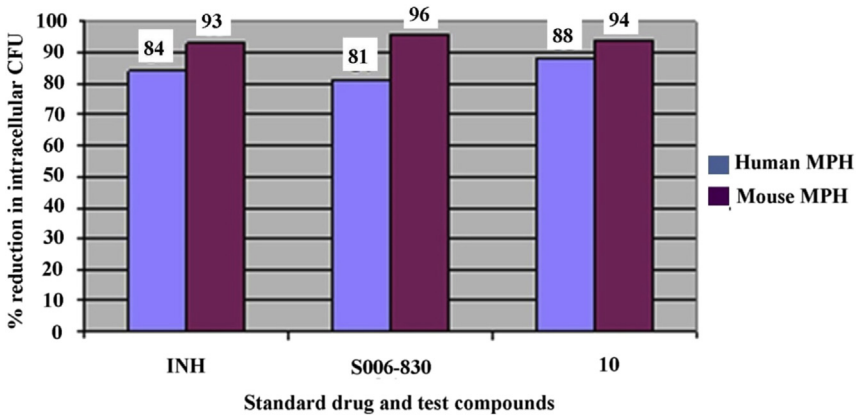


Fig. 4. Compounds **10** and **S006-830** were also able to kill intracellular *M. tuberculosis* H37Rv in human monocyte derived macrophages in a manner similar to the standard drug INH (corresponding mouse macrophage data are shown for comparison).

One possible reason for this is that drug metabolism correlates better with body surface area than body weight. The ratio of body surface area to body weight decreases sharply with increasing body weight. Thus, for most drugs the equipotent dosage is 12 times larger in the mouse than in humans. This assay was performed on outbred Swiss mice (obtained from CDRI-LAD) infected with *M. tuberculosis* $H37R_v$ strain. All four *in vitro* and *ex vivo* active compounds **9, 10, 26** and **S006-830** were evaluated for their *in vivo* efficacy. Swiss mice were infected intravenously with *M. tuberculosis* $H37R_V$ (10^7 CFU/mouse). The experimental groups received daily oral doses of test compounds (100 mg/kg body weight, for 28 days) dissolved in corn oil. Drug-treated control groups received INH (25 mg/kg body weight) or EMB (10 mg/kg body weight) or PZA (150 mg/kg body weight) which were dissolved in water. Untreated control groups received only the vehicles (corn oil or water). Mice were observed up to day-40 for determination of percentage survivors and mean survival time (MST). Maximum survivors (42% in treated and 0% in untreated) were seen in the **S006-830** treated groups, followed by **10** and **26** treated groups. No survivors were left in **9** treated mice. The highest enhancement in MST (7 days, $p < 0.05$) was also seen with **S006-830**. All dead animals were autopsied and their lungs were found full of typical TB lesions. 'Impression smears' of the cut surface of lungs were microscopically examined after staining with Ziehl-Neelsen stain and found full of Acid Fast Bacilli (AFB) confirming that the deaths were due to *M. tuberculosis* infection (Fig. 5).

Fig. 6 shows results of treatment with **S006-830** and the anti-TB drugs-ethambutol (EMB) and pyrazinamide (PZA) in terms of viable bacilli (CFU) in the lungs of mice. **S006-830** treated mice showed CFU count of 2.2×10^7 (mean of 3 animals) which was approximately 15 fold lesser than that of the untreated mice (31×10^7), and comparable with the efficacies of EMB (1.27×10^7) and PZA (1.9×10^7). Based on the result of this and prior studies, compound **S006-830** was chosen as the lead molecule and taken forward for the further assessment.

2.2.4. Minimum bactericidal concentration (MBC) determination of the lead compound 36 (S006-830) through in vitro and ex vivo studies and potency assessment on multi-drug and single-drug resistant M. tuberculosis

These experiments were aimed at determining the capacity of test compounds to kill the bacilli, in addition to inhibiting their multiplication. The assays were thus designed to record killing of the seed culture (inoculum) and in principle, if a compound in *in vitro* study reduces the CFU of the inoculum; it can be considered as bactericidal and corresponding concentration as its MBC. If a compound does not kill the inoculum but only prevents its multiplication (i.e., reduction in day-14 CFU), it can be considered as bacteriostatic (as per method described in the experimental section). For *ex vivo*, if a compound reduces the day-0 CFU (as per method described in the experimental section) of macrophages infected with *M. tuberculosis* it can be considered as bactericidal. If it reduces only the day-5 CFU of such macrophages, it can be considered as bacteriostatic [15]. The lead compound **36** (**S006830**) produced 100% killing (zero CFU) *in vitro* and 75% killing (1200 CFU) *ex vivo* of the inoculum of *M. tuberculosis* $H37R_v$. The bactericidal potency was comparable with standard drug RFM (Fig. 7). S006-830 also showed potent bactericidal activity against multi-drug resistant (i.e., resistant to RFM and INH) and single-drug resistant (to RFM) clinical isolates of *M. tuberculosis* (Fig. 8).

Further, in order to develop S006-830 as identified lead, pharmacokinetics (PK) studies were conducted at Pharmacokinetics & Metabolism Division of our institute CSIR-CDRI using newly developed and validated methods on HPLC and LC-MS/MS for quantitative estimation in rat plasma and demonstrated good PK properties with fast intestinal absorption, peak plasma concentration one hour post oral dose, optimum elimination half-life of 9–13 h, plasma protein binding (PPB) of ~60%, mean residence time of around 18–20 h and favourable bioavailability in the range of 45–50%. As the plasma protein binding was not too high, it will lead to tissue redistribution as well as fast clearance of the unbound fraction of S006-830 from the body. Further, S006-830 was found stable in rat plasma under various operating conditions [9,10].

S006-830 is racemic and after separation through chiral column chromatography, we are separately evaluating the anti-TB activity of both enantiomers and results will be communicated in future.

3. Conclusion

While it was evident from our earlier report that TRSMs can have significant potential against *M. tuberculosis*, this series led us to identify a highly potent and viable lead S006-830. Collectively, from earlier reported compounds [7,8] and the current work, a preliminary structure activity relationship could be drawn and directed to the subsequent consideration: (a) presence of benzene ring with amino alkyl chains is important for potent anti-tubercular activity; (b) TRSMs without any substituent on benzene ring (**5**–**8**) showed moderate anti-tubercular activity; (c) Among TRSMs with electron withdrawing group, *para*-substituted chloro (**9, 10**) containing compounds showed promising *in vitro* activity than their *meta* analogues **11**–**16**. On the other hand, TRSMs with electron donating group i.e., methoxy and thiomethoxy at *para* position of

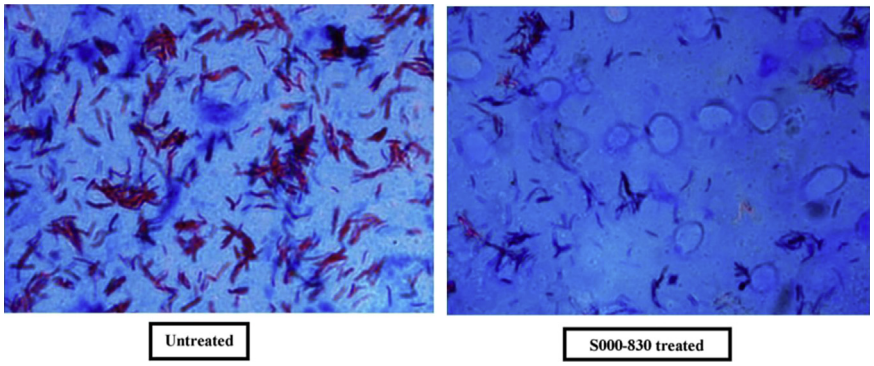


Fig. 5. Ziehl–Neelsen staining of lung tissue smears of mice untreated and treated with **S006-830**.

benzene ring **26, 36** showed good anti-tubercular activities than *ortho* isomers **27**–**35**. Anti-tubercular activity of four TRSMs, which showed MIC of <6.25 mg/L against *M. tuberculosis* $H37R_v$ and lacked cytotoxicity towards mammalian cells showed >90% inhibition in multiplication of *M. tuberculosis* within mouse and human macrophages (*ex vivo*), which was comparable to *ex vivo* efficacy of standard TB drugs-isoniazid, rifampicin and pyrazinamide. The lead molecule S006-830 brought approximately 15 fold reduction in the viable bacilli (CFUs) in lungs of the infected mice, which was comparable to the *in vivo* efficacy of ethambutol and pyrazinamide. **S006-830** with noticeable microbiological profile and encouraging PK properties can be considered as potential lead against *M. tuberculosis*.

4. Experimental section

4.1. Chemistry

4.1.1. General methods

All reagents and solvents were purchased from commercial sources and used without further purification. Organic solvents were dried by standard methods. Analytical TLC was performed using 2.5 × 5 cm aluminium plates coated with a 0.25 mm thickness of silica gel (60F-254), visualization was accomplished with iodine and under UV lamp. The spots on TLC were also visualized by warming ceric sulphate (2% $CeSO_4$ in 2 N H_2SO_4) sprayed plates in hot plate or in oven at about 100 °C. Column chromatography was performed using silica gel (60–120 and 100–200 mesh). ^{1}H NMR spectra were recorded on 200 and 300 MHz spectrometer in $CDCl_3$ (all signals are reported in ppm with the internal chloroform signal at 7.26 ppm as standard) at 25 °C. ^{13}C NMR spectra were recorded on 50 and 75 MHz spectrometer in $CDCl_3$ (all signals are reported in ppm with the internal chloroform signal at 77.00 ppm as standard) at 25 °C. In a few cases tetramethylsilane (TMS) at 0.00 ppm was used as the reference standard. Coupling constants (*J* values) are given in hertz (Hz). ^{1}H NMR splitting patterns are designated as singlet (s), doublet (d), double doublet (dd), triplet (t), quartet (q) or multiplet (m). IR spectra were recorded using a FTIR spectrophotometer in cm^{-1}.

*4.1.1.1. General procedure for the preparation of heteroarylcarbinols **(3)** through Grignard reaction.* To a suspension of Mg (1.50 g, 61.7 mmol) in dry THF (40 mL) was added drop wise a solution of 4-bromoanisole (6.52 mL, 52.1 mmol) in dry THF (40 mL). After stirring the mixture for 30 min a solution of thiophenylcarbaldehyde (4.21 mL, 45 mmol) in dry THF (30 mL) was added drop wise and the resulting solution was allowed to stir for further 30 min. After quenching by adding a saturated solution of NH_4Cl (20 mL), the reaction mixture was extracted with ethyl acetate (100 mL), washed with water (100 mL), brine (2 × 50 mL) and then dried over Na_2SO_4. The organic layer was dried under reduced pressure. The crude product was purified by column chromatography.

4.1.1.2. General procedure for the preparation of thiophene containing trisubstituted methanes through Friedel Crafts alkylation reaction. Phenol, benzene, 1-2 drops of sulphuric acid along with the carbinol **3** was refluxed at 60–75 °C for 2–3 h. Completion of reaction was confirmed by checking TLC by which compound **4** was obtained. After completion, reaction was quenched with water, reaction mixture was extracted with ethyl acetate (100 mL), washed with water and brine and dried over Na_2SO_4. Solvent was removed under reduced pressure and purified by column chromatography.

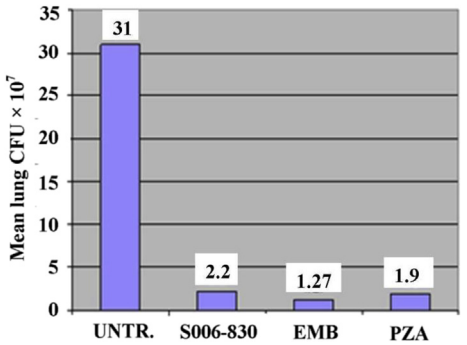


Fig. 6. CFU in the lungs of mice treated with the **S006-830** and standard drugs ethambutol (EMB) and pyrazinamide (PZA). UNTR stands for untreated.

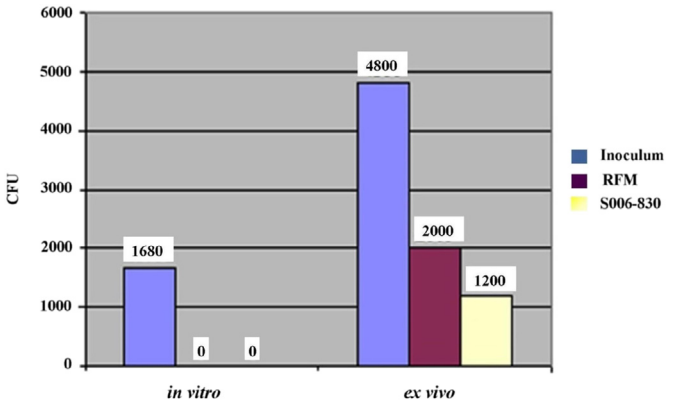


Fig. 7. *In vitro* and *ex vivo* bactericidal property of **36 (S006-830)** in comparison with rifampicin (RFM).

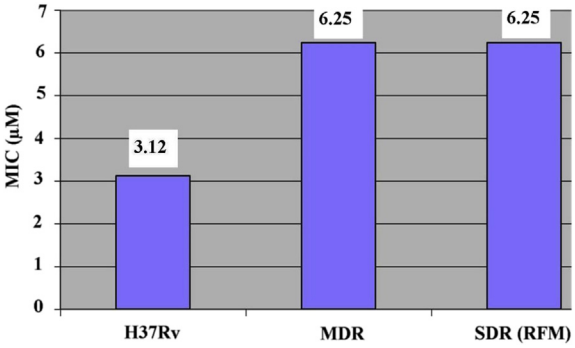


Fig. 8. Activity of **36** (**S006-830**) against sensitive ($H37R_V$), single- and multi-drug resistant *M. tuberculosis*.

*4.1.1.3. Diethyl-{2-[4-(phenyl-thiophen-2-yl-methyl)-phenoxy]-ethyl}-amine **(5)**.* Compound **4** (0.2 g, 0.8 mmol) refluxed with K_2CO_3 (0.2 g, 1.9 mmol), 1-(2-chloroethyl)-diethyl amine hydrochloride (0.2 g, 0.7 mmol) in acetone furnished **5** (0.2 g, 72.8%) as brown viscous oil. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.18–7.15 (m, 8H), 6.91–6.78 (m, 3H), 6.64–6.63 (m, 1H), 5.59 (s, 1H), 4.02 (t, J = 6.22, 2H), 2.87 (t, J = 6.2, 2H), 2.65 (q, J = 7.1, 4H), 1.08 (t, J = 7.0, 6H); ^{13}C NMR ($CDCl_3$, 50 MHz): δ 155.8, 147.9, 143.7, 137.7, 130.1, 129.8, 128.7 (2C), 128.4 (2C), 128.3 (2C), 126.7, 126.6, 126.3, 124.6, 114.5 (2C), 65.6, 51.2, 48.8, 47.7, 11.2; IR (Neat): 3019, 2928, 2855, 2362, 1612, 1509, 1217, 761 cm^{-1}; MS (ESI): m/z 366 $(M+H)^+$; Anal. Calcd for $C_{23}H_{27}NOS$: C, 75.57; H, 7.45; N, 3.83. Found: C, 75.61; H, 7.52; N, 3.72.

*4.1.1.4. 1-{2-[4-(Phenyl-thiophen-2-yl-methyl)-phenoxy]-ethyl}-piperidine **(6)**.* Compound **4** (200 mg, 0.75 mmol) refluxed with K_2CO_3 (259.42 mg, 1.87 mmol), 1-(2-chloroethyl)-piperidine hydrochloride (138.23 mg, 0.75 mmol) in acetone furnished **6** (203.8 mg, 71.9%) as brown viscous oil. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.28–7.15 (m, 6H), 7.09–7.06 (m, 2H), 6.90–6.87 (m, 1H), 6.81–6.78 (m, 2H), 6.64–6.63 (m, 1H), 5.58 (s, 1H), 4.07 (t, J = 6.0, 2H), 2.76 (t, J = 6.0, 2H), 2.52 (t, J = 4.9, 4H), 1.65–1.58 (m, 4H), 1.48–1.42 (m, 2H); ^{13}C NMR ($CDCl_3$, 50 MHz): δ 157.2, 156.6, 148.5, 136.1, 132.8, 129.8 (2C), 129.6 (2C), 127.7, 126.3, 126.0, 123.9, 120.4, 114.1, 110.5, 96.1, 65.5, 57.8, 55.5, 54.9, 43.8, 25.6, 24.0; IR (Neat): 3020, 2936, 2362, 1610, 1508, 1217, 761 cm^{-1}; MS (ESI): m/z 378 $(M+H)^+$; Anal. Calcd for $C_{24}H_{27}NOS$: C, 76.35; H, 7.21; N, 3.71. Found: C, 76.40; H, 7.15; N, 3.69.

*4.1.1.5. 1-{2-[4-(Phenyl-thiophen-2-yl-methyl)-phenoxy]-ethyl}-azepane **(7)**.* Compound **4** (200 mg, 0.75 mmol) refluxed with K_2CO_3 (259.42 mg, 1.87 mmol), 1-(2-Chloro-ethyl)-azepane hydrochloride (148.58 mg, 0.75 mmol) in acetone furnished **7** (220 mg, 75%) as brown viscous oil. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.21–7.06 (m, 8H), 6.91–6.78 (m, 3H), 6.64–6.63 (m, 1H), 5.59 (s, 1H), 4.07 (t, J = 6.0, 2H), 2.97 (t, J = 6.1, 2H), 2.81 (t, J = 5.6, 4H), 1.69–1.61 (m, 8H); ^{13}C NMR ($CDCl_3$, 50 MHz): δ 157.5, 148.4, 144.1, 136.2, 129.8 (2C), 128.7 (2C), 128.4 (2C), 126.6, 126.5, 126.2, 124.4, 114.4 (2C), 65.7, 57.8, 54.9 (2C), 51.3, 25.7 (3C), 24.1; IR (Neat): 3019, 2938, 2362, 1610, 1509, 1218, 763 cm^{-1}; MS (ESI): m/z 392 $(M+H)^+$; Anal. Calcd for $C_{25}H_{29}NOS$: C, 76.68; H, 7.46; N, 3.58. Found: C, 76.72; H, 7.39; N, 3.61.

*4.1.1.6. 4-{2-[4-(Phenyl-thiophen-2-yl-methyl)-phenoxy]-ethyl}morpholine **(8)**.* Compound **4** (200 mg, 0.75 mmol) refluxed with K_2CO_3 (259.42 mg, 1.87 mmol), 4-(2-Chloro-ethyl)-morpholine hydrochloride (258.45 mg, 0.75 mmol) in acetone furnished **8** (205.1 mg, 72%) as brown viscous oil. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.29–7.15 (m, 6H), 7.10–7.06 (m, 2H), 6.90–6.78 (m, 3H), 6.64–6.63 (m, 1H), 5.59 (s, 1H), 4.06 (t, J = 5.7, 2H), 3.70 (t, J = 4.4, 4H), 2.77 (t, J = 5.6, 2H), 2.55 (t, J = 4.6, 4H); ^{13}C NMR ($CDCl_3$, 50 MHz): δ 157.4, 148.3, 144.0, 136.3, 129.8, 128.7, 128.7 (2C), 128.5 (2C), 128.4, 126.6, 126.5, 126.2, 124.4, 114.4 (2C), 66.7, 65.5, 57.6, 54.0 (2C), 51.3; IR (Neat): 3018, 2927, 2862, 1609, 1509, 1217, 753 cm^{-1}; MS (ESI): m/z 378 $(M-H)^+$, 379 (M^+), 380 $(M+H)^+$; Anal. Calcd for $C_{23}H_{25}NO_2S$: C, 72.79; H, 6.64; N, 3.69. Found: C, 72.82; H, 6.70; N, 3.55.

*4.1.1.7. 1-(2-{4-[(4-Chloro-phenyl)-thiophen-2-yl-methyl]-phenoxy}-ethyl)-piperidine **(9)**.* Compound **4** p-Cl substituted at Ar[1] (0.20 g, 0.667 mmol) refluxed with K_2CO_3 (0.280 g, 2.00 mmol), 1-(2-Chloroethyl)-piperidine hydrochloride (0.135 g, 0.734 mmol) in acetone furnished **9** (0.205 g, 75%) as wine red viscous liquid. ^{1}H NMR (200 MHz, $CDCl_3$): δ 7.15–6.94 (m, 7H), 6.82–6.52 (m, 4H), 5.47 (s, 1H), 3.99 (t, J = 4.0, 2H), 2.67 (t, J = 4.0, 2H), 2.45–2.41 (m, 4H), 1.57–1.37 (m, 6H); ^{13}C NMR ($CDCl_3$, 50 MHz): δ 158.1, 148.1, 143.0, 135.8, 132.8, 130.4 (2C), 130.0 (2C), 128.9 (2C), 126.9, 126.6, 125.0, 114.8 (2C), 77.4, 66.2, 58.3, 55.4, 51.0, 30.1, 26.3, 24.6; IR (Neat) 3438, 2990, 1712, 1623, 775, 664 cm^{-1}; MS (ESI): m/z 412 $(M+H)^+$; Anal. Calcd for $C_{24}H_{26}ClNOS$: C, 69.97; H, 6.36; N, 3.40. Found: C, 70.04; H, 6.34; N, 3.46.

*4.1.1.8. (2-{4-[(4-Chloro-phenyl)-thiophen-2-yl-methyl]-phenoxy}-ethyl)-diisopropyl-amine **(10)**.* Compound **4** p-Cl substituted at Ar[1] (0.20 g, 0.67 mmol) refluxed K_2CO_3 (0.28 g, 2.00 mmol), (2-Chloroethyl)-diisopropyl-amine hydrochloride (0.15 g, 0.74 mmol) in acetone furnished **10** as yellow viscous oil (0.21 g, 73%). ^{1}H NMR (200 MHz, $CDCl_3$): δ 7.29–7.19 (m, 4H), 7.14–7.05 (m, 3H), 6.94–6.64 (m, 4H), 5.58 (s, 1H), 3.87 (t, J = 7.4, 2H), 3.07–3.00 (m, 2H), 2.80 (t, J = 7.4, 2H), 1.03 (d, J = 6.5, 12H); ^{13}C NMR ($CDCl_3$, 50 MHz): δ 156.6, 146.5, 141.4, 134.0, 131.1, 128.8 (2C), 128.4 (2C), 127.2 (2C), 125.3 (2C), 124.9, 123.3, 113.1 (2C), 67.9, 49.4, 48.2, 43.1, 19.5 (4C); IR (Neat): 3312, 2967, 1602, 1446, 1353, 1023, 759 cm^{-1}; MS (ESI): m/z 428 $(M+H)^+$; Anal. Calcd for $C_{25}H_{30}ClNOS$: C, 70.15; H, 7.06; N, 3.27. Found: C, 70.13; H, 7.13; N, 3.31.

*4.1.1.9. 2-(4-((3-Chlorophenyl) (thiophen-2-yl)methyl)phenoxy)-N,N-dimethylethanamine **(11)**.* Yield, 72%; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.19–7.18 (m 4H), 7.09–7.07 (m, 3H), 6.93–6.90 (m, 1H), 6.86–6.84 (m, 2H), 6.66–6.65 (m, 1H), 5.57 (s, 1H), 4.04 (t, J = 5.7, 2H), 2.72 (t, J = 5.7, 2H), 2.33 (s, 6H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 157.6, 147.3, 146.0, 135.2, 134.1, 129.6, 129.5 (2C), 128.7, 126.9, 126.7, 126.5, 126.3, 124.6, 114.3 (2C), 65.7, 58.1, 50.8, 45.8 (2C); IR (Neat) 3471, 2980, 1748, 1643, 768 cm^{-1}; MS (ESI) m/z 372 $(M+H)^+$; Anal. Calcd for $C_{21}H_{22}ClNOS$: C, 67.82; H, 5.96; Cl, 9.53; N, 3.77. Found: C, 67.77; H, 5.92; N, 3.71.

*4.1.1.10. 2-(4-((3-Chlorophenyl) (thiophen-2-yl)methyl)phenoxy)-N,N-diethylethanamine **(12)**.* Yield, 75%; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.21–7.18 (m, 4H), 7.09–7.07 (m, 3H), 6.94–6.91 (m, 1H), 6.85–6.82 (m, 2H), 6.67–6.66 (m, 1H), 5.58 (s, 1H), 4.02 (t, J = 6.4, 2H), 2.86 (t, J = 6.4, 2H), 2.63 (q, J = 7.1, 4H), 1.06 (t, J = 7.1, 6H); ^{13}C NMR (50 MHz, $CDCl_3$): δ 157.4, 146.1, 135.5, 134.2, 129.8, 129.6 (2C), 128.8 (2C), 126.9, 126.8, 126.6, 126.4, 124.7, 114.4 (2C), 65.7, 51.5, 50.9, 47.6 (2C), 11.1 (2C); IR (Neat) 3433, 2985, 1747, 1611, 779 cm^{-1}; MS (ESI) m/z 400 $(M+H)^+$; Anal. Calcd for $C_{23}H_{26}ClNOS$: C, 69.07; H, 6.55; N, 3.50. Found: C, 69.13; H, 6.64; N, 3.55.

*4.1.1.11. 1-(2-(4-((3-Chlorophenyl) (thiophen-2-yl)methyl)phenoxy) ethyl) pyrrolidine **(13)**.* Yield, 68%; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.19–7.18 (m, 4H), 7.10–7.07 (m, 3H), 6.93–6.90 (m, 1H), 6.86–6.83 (m, 2H), 6.66–6.65 (m, 1H), 5.57 (s, 1H), 4.12 (t, J = 5.8, 2H), 2.94 (t, J = 5.8, 2H), 2.69 (s, 4H), 1.82 (s, 4H); ^{13}C NMR (50 MHz, $CDCl_3$): δ 157.5, 147.2, 146.0, 135.3, 134.1, 129.6, 129.5, 128.7 (2C),

126.8, 126.7, 126.5, 126.2, 124.6, 114.4 (2C), 66.5, 54.8, 54.5 (2C), 50.8, 23.3 (2C); IR (Neat) 3451, 2962, 1745, 1611, 871 cm^{-1}; MS (ESI) *m/z* 398 $(M+H)^+$; Anal. Calcd for $C_{23}H_{24}ClNOS$: C, 69.42; H, 6.08; N, 3.52. Found: C, 69.33; H, 6.04; N, 3.55.

4.1.1.12. 1-(2-(4-((3-Chlorophenyl) (thiophen-2-yl)methyl)phenoxy) ethyl) piperidine ***(14)***. Yield, 79%; ^1H NMR (300 MHz, $CDCl_3$) δ 7.19–7.17 (m, 4H), 7.09–7.06 (m, 3H), 6.93–6.90 (m, 1H), 6.85–6.82 (m, 2H), 6.67–6.66 (m, 1H), 5.57 (s, 1H), 4.08 (t, $J = 6.6$, 2H), 2.77 (t, $J = 5.9$, 2H), 2.53–2.50 (m, 4H), 1.64–1.57 (m, 4H), 1.47–1.43 (m, 2H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 157.5, 147.2, 146.0, 135.1, 134.0, 129.6, 129.4, 128.6, 126.8 (2C), 126.7, 126.5, 126.2, 124.6, 114.3 (2C), 65.6, 57.7, 54.8 (2C), 50.8, 25.6 (2C), 23.9; MS (ESI) *m/z* 412 $(M+H)^+$; Anal. Calcd for $C_{24}H_{26}ClNOS$: C, 69.97; H, 6.36; N, 3.40. Found: C, 69.89; H, 6.34; N, 3.45.

4.1.1.13. N-(2-(4-((3-chlorophenyl) (thiophen-2-yl)methyl)phenoxy) ethyl)-N-isopropylpropan-2-amine ***(15)***. Yield, 63%; ^1H NMR (300 MHz, $CDCl_3$) δ 7.19–7.18 (m, 4H), 7.09–7.06 (m, 3H), 6.92–6.89 (m, 1H), 6.84–6.81 (m, 2H), 6.66 (d, $J = 3.3$, 1H), 5.57 (s, 1H), 3.87 (t, $J = 7.4$, 2H), 3.07–2.98 (m, 2H), 2.80 (t, $J = 7.5$, 2H), 1.02 (d, $J = 6.5$, 12H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 157.4, 147.4, 146.1, 135.0, 134.2, 129.6, 129.5, 128.8, 126.9 (2C), 126.8, 126.6, 126.3, 124.6, 114.3 (2C), 69.2, 50.9, 49.6 (2C), 44.4, 20.8 (4C); IR (Neat) 3432, 2990, 1742, 1631 cm^{-1}; MS (ESI) *m/z* 428 $(M+H)^+$; Anal. Calcd for $C_{25}H_{30}ClNOS$: C, 70.15; H, 7.06; N, 3.27. Found: C, 69.99; H, 7.14; N, 3.30.

4.1.1.14. 4-(2-(4-((3-Chlorophenyl) (thiophen-2-yl)methyl)phenoxy) ethyl) morpholine ***(16)***. Yield, 68%; ^1H NMR (300 MHz, $CDCl_3$) δ 7.20–7.18 (m, 4H), 7.10–7.08 (m, 3H), 6.94–6.92 (m, 1H), 6.85–6.83 (m, 2H), 6.67–6.66 (m, 1H), 5.58 (s, 1H), 4.09 (t, $J = 4.5$, 2H), 3.72 (t, $J = 3.5$, 4H), 2.79 (t, $J = 4.4$, 2H), 2.57 (d, $J = 3.0$, 4H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 157.6, 147.3, 146.1, 135.4, 134.2, 129.7, 129.5, 128.8, 126.9 (2C), 126.8, 126.6, 126.3, 124.7, 114.5 (2C), 66.9 (2C), 65.7, 57.6, 54.0 (2C), 50.9; IR (Neat) 3435, 2980, 1732, 1630 cm^{-1}. MS (ESI) *m/z* 414 $(M+H)^+$; Anal. Calcd for $C_{23}H_{24}ClNO_2S$: C, 66.73; H, 5.84; N, 3.38. Found: C, 66.79; H, 5.88; N, 3.31.

4.1.1.15. 2-(4-((2-Fluorophenyl) (thiophen-2-yl)methyl)phenoxy)-N,N-dimethylethanamine ***(17)***. Isolated as light brownish oily liquid (yield 71%). ^1H NMR (300 MHz, $CDCl_3$): δ 7.25–7.22 (m, 1H), 7.21–7.12 (m, 1H), 7.09–7.02 (m, 5H), 6.94–6.91 (m, 1H), 6.87–6.67 (m, 2H), 6.66 (d, $J = 2.5$, 1H), 5.93 (s, 1H), 4.05 (t, $J = 5.7$, 2H), 2.73 (t, $J = 5.7$, 2H), 2.33 (s, 6H). ^{13}C NMR (50 MHz, $CDCl_3$): δ 162.7, 157.6, 146.9, 134.8, 130.1, 129.6, 128.5, 126.6 (2C), 126.3, 124.5, 124.0, 115.5, 115.1, 114.4 (2C), 65.8, 58.2, 45.8 (2C), 43.7; IR (Neat): 3432, 2904, 1611, 1445, 1249, 1176, 1023, 823, 778, 661 cm^{-1}; MS (ESI): *m/z* 356 $(M+H)^+$, Anal. Calcd for $C_{21}H_{22}FNOS$: C, 70.96; H, 6.24; N, 3.94. Found: C, 70.91; H, 6.23; N, 3.97.

4.1.1.16. N, N-diethyl-2-(4-((2-fluorophenyl) (thiophen-2-yl)methyl) phenoxy) ethanamine ***(18)***. Isolated as light blackish oily liquid (yield 64%). ^1H NMR (300 MHz, $CDCl_3$): δ 7.24–7.12 (m, 2H), 7.09–7.01 (m, 5H), 6.93–6.90 (m, 1H), 6.84–6.81 (m, 2H), 6.66 (d, $J = 3.4$, 1H), 5.93 (s, 1H), 4.04 (t, $J = 6.3$, 2H), 2.88 (t, $J = 6.3$, 2H), 2.65 (q, $J = 7.1$, 4H), 1.07 (d, $J = 7.1$, 6H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 162.7, 157.6, 146.9, 134.7, 130.1, 129.6, 128.4, 128.3, 126.5, 126.2 (2C), 124.5, 123.9, 115.5, 114.3 (2C), 66.2, 51.7, 47.7 (2C), 43.7, 11.6 (2C); IR (Neat): 3433, 2904, 1617, 1445, 1254, 1176, 1033, 823, 779, 662 cm^{-1}; MS (ESI): *m/z* 384 $(M+H)^+$, Anal. Calcd for $C_{23}H_{26}FNOS$: C, 72.03; H, 6.83; N, 3.65. Found: C, 72.09; H, 6.80; N, 3.67.

4.1.1.17. 1-(2-(4-((2-Fluorophenyl) (thiophen-2-yl)methyl)phenoxy) ethyl) pyrrolidine ***(19)***. Isolated as brownish oily liquid (yield 68%). ^1H NMR (300 MHz, $CDCl_3$): δ 7.25–7.12 (m, 2H), 7.09–6.99 (m, 5H), 6.94–6.91 (m, 1H), 6.86–6.83 (m, 2H), 6.66 (d, $J = 0.9$, 1H), 5.93 (s, 1H), 4.09 (t, $J = 5.9$, 2H), 2.90 (t, $J = 5.9$, 2H), 2.63 (s, 4H), 1.80 (d, $J = 3.8$, 4H). ^{13}C NMR (50 MHz, $CDCl_3$): δ 157.7, 146.9, 139.2, 134.7, 130.1, 129.6, 128.5, 128.3, 126.5 (2C), 126.3, 124.5, 124.0, 123.9, 115.5, 114.4 (2C), 66.8, 54.6 (2C), 43.7, 23.4 (2C); IR (Neat): 3433, 2909, 1617, 1445, 1235, 1176, 1043, 823, 779, 660 cm^{-1}; MS (ESI): *m/z* 382 $(M+H)^+$, Anal. Calcd for $C_{23}H_{24}FNOS$: C, 72.41; H, 6.34; N, 3.67. Found: C, 72.42; H, 6.39; N, 3.71.

4.1.1.18. 1-(2-(4-((2-Fluorophenyl) (thiophen-2-yl)methyl)phenoxy) ethyl) piperidine ***(20)***. Isolated as light brownish oily liquid (yield 75%). ^1H NMR (300 MHz, $CDCl_3$): δ 7.24–7.12 (m, 2H), 7.09–6.98 (m, 5H), 6.93–6.90 (m, 1H), 6.84–6.81 (m, 2H), 6.66 (d, $J = 3.4$, 1H), 5.92 (s, 1H), 4.09 (t, $J = 6.0$, 2H), 2.78 (t, $J = 6.0$, 2H), 2.52 (t, $J = 4.6$, 4H), 1.64–1.57 (m, 4H), 1.47–1.40 (m, 2H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 159.1, 157.6, 146.9, 134.8, 131.3, 129.7 (2C), 128.5, 128.4, 126.6, 124.5, 115.4, 114.4 (2C), 65.6, 57.8, 54.9 (2C), 43.8, 43.7, 37.9, 25.7 (2C), 24.0; IR (Neat): 3423, 2914, 1617, 1425, 1254, 1176, 1031, 823, 774, 661 cm^{-1}; MS (ESI): *m/z* 396 $(M+H)^+$, Anal. Calcd for $C_{24}H_{26}FNOS$: C, 72.88; H, 6.63; N, 3.54. Found: C, 72.83; H, 6.57; N, 3.59.

4.1.1.19. N-(2-(4-((2-fluorophenyl) (thiophen-2-yl)methyl)phenoxy) ethyl)-N-isopropylpropan-2-amine ***(21)***. Isolated as light brownish oily liquid (yield 72%). ^1H NMR (300 MHz, $CDCl_3$): δ 7.22–7.11 (m, 2H), 7.08–6.97 (m, 5H), 6.92–6.89 (m, 1H), 6.83–6.81 (m, 2H), 6.66 (d, $J = 2.5$, 1H), 5.92 (s, 1H), 3.86 (t, $J = 7.3$, 2H), 3.07–2.98 (m, 2H), 2.80 (t, $J = 7.3$, 2H), 1.02 (d, $J = 6.5$, 12H).^{13}C NMR (75 MHz, $CDCl_3$): δ 162.7, 157.8, 147.0, 134.4, 130.1, 129.6, 128.4, 126.5, 126.2 (2C), 124.4, 123.9, 115.5, 115.0, 114.2 (2C), 69.1, 49.6 (2C), 44.4, 43.7, 20.8 (4C); IR (Neat): 3432, 2924, 1621, 1458, 1249, 1166, 1033, 833, 768, 663 cm^{-1}; MS (ESI): *m/z* 412 $(M+H)^+$, Anal. Calcd for $C_{25}H_{30}FNOS$: C, 72.96; H, 7.35; N, 3.40. Found: C, 72.92; H, 7.27; N, 3.46.

4.1.1.20. 4-(2-(4-((2-Fluorophenyl) (thiophen-2-yl)methyl)phenoxy) ethyl) morpholine ***(22)***. Isolated as brownish oily liquid (yield 71%). ^1H NMR (300 MHz, $CDCl_3$): δ 7.24–7.21 (m, 1H), 7.19–7.12 (m, 1H), 7.09–6.98 (m, 5H), 6.93–6.90 (m, 1H), 6.84–6.81 (m, 2H), 6.66 (d, $J = 3.3$, 1H), 5.92 (s, 1H), 4.09 (s, 2H), 3.72 (t, $J = 4.5$, 4H), 2.80 (s, 2H), 2.60 (d, $J = 4.5$, 4H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 161.5, 159.1, 146.6, 135.0, 129.7, 128.5 (2C), 128.4 (2C), 126.5 (2C), 124.6 (2C), 115.2, 114.4 (2C), 66.8 (2C), 65.6, 57.6, 54.0 (2C), 37.9; IR (Neat): 3453, 2904, 1617, 1445, 1224, 1176, 1033, 823, 770, 662 cm^{-1}; MS (ESI): *m/z* 398 $(M+H)^+$, Anal. Calcd for $C_{23}H_{24}FNO_2S$: C, 69.49; H, 6.09; N, 3.52. Found: C, 69.51; H, 6.13; N, 3.48.

4.1.1.21. 2-(4-((3-Fluorophenyl) (thiophen-2-yl)methyl)phenoxy)-N,N-dimethylethanamine ***(23)***. Colourless oil; yield, 78%; ^1H NMR (300 MHz, $CDCl_3$) δ 7.20–7.17 (m, 3H), 7.00–6.86 (m, 7H), 6.65 (s, 1H), 6.07 (s, 1H), 4.02–3.96 (m, 2H), 2.55 (t, $J = 5.7$, 2H), 2.23 (s, 6H); ^{13}C NMR (75 MHz, $CDCl_3$): δ *157.6*, *147.3*, *146.0*, *135.2*, *129.5*, *128.7* (2C), *126.9*, *126.7*, *126.5*, *125.9*, *125.4*, *125.0*, *124.6*, *114.3* (2C), *65.7*, *58.1*, *50.8*, *45.8* (2C); IR (Neat): 3015, 2871, 2121, 1580, 1434, 1251, 1013, 729 cm^{-1}; MS (ESI): *m/z* 356 $[M+H]^{+\cdot}$; Anal. Calcd for $C_{21}H_{22}FNOS$: C, 70.96; H, 6.24; N, 3.94. Found: C, 70.90; H, 6.30; N, 3.90.

4.1.1.22. N, N-diethyl-2-(4-((3-fluorophenyl) (thiophen-2-yl)methyl) phenoxy) ethanamine ***(24)***. Colourless oil; yield, 75%; ^1H NMR (300 MHz, $CDCl_3$) δ 7.14–7.10 (m, 3H), 7.02–6.99 (m, 2H), 6.86–6.59 (m, 5H), 6.59 (s, 1H), 5.52 (s, 1H), 3.94 (t, $J = 6.3$, 2H), 2.78 (t, $J = 6.3$, 2H), 2.58–2.51 (m, 4H), 0.98 (t, $J = 2.3$, 6H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 164.5, 157.7, 147.5, 135.3, 129.7, 126.6, 126.3 (2C), 124.6 (2C), 115.8, 115.5, 114.4 (2C), 113.6, 113.3, 66.4, 51.7, 50.9, 47.8 (2C), 11.8 (2C); IR (neat): 3076, 2811, 2190, 1581, 1446, 1242, 1039,

777 cm^{-1}; MS (ESI): *m/z* 384 $(M+H)^+$; Anal. Calcd for $C_{23}H_{26}FNOS$: C, 72.03; H, 6.83; N, 3.65. Found: C, 72.06; H, 6.89; N, 3.71.

4.1.1.23. 1-(2-(4-((3-Fluorophenyl) (thiophen-2-yl)methyl)phenoxy) ethyl) piperidine ***(25)****.* Colourless oil; yield, 77%; ^1H NMR (300 MHz, $CDCl_3$) δ 7.11–7.06 (m, 3H), 6.92–6.56 (m, 7H), 6.55 (s, 1H), 5.98 (s, 1H), 3.97–3.92 (m, 2H), 2.53 (t, *J* = 5.7, 2H), 2.28 (t, *J* = 4.3, 4H), 1.45–1.17 (m, 6H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 164.0, 155.8, 147.0, 139.2, 131.9, 129.5, 128.2, 126.5 (2C), 126.4 (2C), 124.3 (2C), 115.9, 115.7, 114.6 (2C), 66.5, 57.8 (2C), 44.8, 25.8 (3C); IR (Neat): 3042, 2862, 2115, 1587, 1491, 1211, 1047, 769 cm^{-1}; MS (ESI): *m/z* 396 $(M+H)^+$; Anal. Calcd for $C_{24}H_{26}FNOS$: C, 72.88; H, 6.63; N, 3.54. Found: C, 72.82; H, 6.69; N, 3.50.

4.1.1.24. N-isopropyl-N-(2-(4-((4-(methylthio)phenyl) (thiophen-2-yl)methyl)phenoxy)ethyl)propan-2-amine ***(26)****.* Compound **4** *p*-SMe substituted at Ar^1 (0.10 g, 0.35 mmol) refluxed with K_2CO_3 (0.135 g, 0.96 mmol), (2-Chloro-ethyl)-diethyl-amine hydrochloride (0.07 g, 0.35 mmol) in acetone furnished **26** as a light yellow viscous oil (0.105 g, 74%); ^1H NMR (200 MHz, $CDCl_3$): δ 7.18–6.99 (m, 7H), 6.86–6.85 (m, 1H), 6.74 (d, *J* = 8.1, 2H), 6.60 (d, *J* = 3.6, 1H), 5.49 (s, 1H), 3.80 (t, *J* = 7.4, 2H), 2.99–2.93 (m, 2H), 2.73 (t, *J* = 7.4, 2H), 2.38 (s, 3H), 0.96 (d, *J* = 6.4, 12H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 158.1, 148.7, 141.6, 136.9, 136.1, 130.1 (2C), 129.6 (2C), 127.0 (2C), 126.6 (2C), 124.8, 114.7 (2C), 69.5, 51.2, 50.1 (2C), 44.9, 21.2 (4C), 16.3; IR (Neat): 2964, 1608, 1505, 1248 cm^{-1}; MS (ESI): *m/z* 440 $(M+H)^+$, Anal. Calcd for $C_{26}H_{33}NOS_2$: C, 71.02; H, 7.57; N, 3.19. Found: C, 71.04; H, 7.63; N, 3.24.

4.1.1.25. Dimethyl-(2-{4-[(2-methylsulfanyl-phenyl)-thiophen-2-yl-methyl]-phenoxy}-ethyl)-amine ***(27)****.* Compound **4** *o*-SMe substituted at Ar^1 (200 mg, 0.64 mmol) refluxed with K_2CO_3 (221.17 mg, 1.6 mmol), 1-(2-chloroethyl)-dimethyl amine hydrochloride (92.20 mg, 0.64 mmol) in acetone furnished **27** (174 mg, 70.8%) as brown viscous oil; ^1H NMR (300 MHz, $CDCl_3$): δ 7.22–7.14 (m, 3H), 7.05–7.01 (m, 4H), 6.89–6.78 (m, 3H), 6.59–6.57 (m, 1H), 6.07 (s, 1H), 4.02 (t, *J* = 5.8, 2H), 2.70 (t, *J* = 5.7, 2H), 2.36 (s, 3H), 2.32 (s, 6H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 157.7, 156.7, 136.1, 129.8 (3C), 127.8, 126.4, 126, 124 (2C), 120 (2C), 114 (2C), 110, 65.8, 58.3, 55.6, 45.8 (2C), 43.8; IR (Neat): 3017, 2926, 2361, 2338, 1609, 1508, 1465, 1217, 1040, 762 cm^{-1}; MS (ESI): *m/z* 384 $(M+H)^+$; Anal. Calcd for $C_{22}H_{25}NOS_2$: C, 68.89; H, 6.57; N, 3.65. Found: C, 68.95; H, 6.62; N, 4.09.

4.1.1.26. Diethyl-(2-{4-[(2-methylsulfanyl-phenyl)-thiophen-2-yl-methyl]-phenoxy}-ethyl)-amine ***(28)****.* Compound **4** *o*-SMe substituted at Ar^1 (200 mg, 0.64 mmol)refluxed with K_2CO_3 (221.17 mg, 1.6 mmol), 1-(2-chloroethyl)-diethyl amine hydrochloride (110.16 mg, 0.64 mmol)in acetone furnished **28** (191.7 mg, 72.7%) as brown viscous oil; ^1H NMR (300 MHz, $CDCl_3$): δ 7.21–7.14 (m, 4H), 7.08–6.99 (m, 4H), 6.89–6.76 (m, 2H), 6.59–6.58 (m, 1H), 6.06 (s, 1H), 4.01 (t, *J* = 6.3, 2H), 2.86 (t, *J* = 6.2, 2H), 2.67–2.60 (m, 4H), 2.36 (s, 3H), 1.07 (t, *J* = 7.0, 6H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 157.4, 137.5, 135.4, 130.1 (2C), 129.1 (2C), 127.3, 126.6 (3C), 125.1 (2C), 124.5, 114.3 (2C), 65.8, 51.5, 47.7, 47.6, 42.3, 16.5, 11.3 (2C); IR (Neat): 2967, 2358, 1617, 1222, 1040, 767 cm^{-1}; MS (ESI): *m/z* 412 $(M+H)^+$, Anal. Calcd for $C_{24}H_{29}NOS_2$: C, 70.03; H, 7.10; N, 3.40. Found: C, 70.10; H, 7.19; N, 3.30.

4.1.1.27. (2-{4-[(2-Methoxy-phenyl)-thiophen-2-yl-methyl]-phenoxy}-ethyl)-dimethyl-amine ***(29)****.* Compound **4** *o*-OMe substituted at Ar^1 (400 mg, 1.349 mmol) refluxed with K_2CO_3 (466.32 mg, 3.374 mmol), 1-(2-chloroethyl)-dimethyl amine hydrochloride (194.32 mg, 1.349 mmol) in acetone furnished **29** (350 mg, 70.5%) as brown viscous oil; ^1H NMR (300 MHz, $CDCl_3$): δ 7.21–6.99 (m, 5H), 6.89–6.78 (m, 5H), 6.61–6.60 (m, 1H), 6.00 (s, 1H), 4.02 (t, *J* = 5.8, 2H), 3.74 (s, 3H), 2.71 (t, *J* = 5.7, 2H), 2.32 (s, 6H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 157.3, 156.7, 136.1, 129.8 (3C), 127.8, 126.4 (2C), 126.0 (2C), 124.0, 120.4, 114.2 (2C), 110.7, 65.5, 58.1, 55.4, 45.6 (2C), 43.8; IR (Neat): 3011, 2933, 26 21, 1608, 1509, 1463, 1243, 1218, 1032, 758 cm^{-1}; MS (ESI): *m/z* 368 $(M+H)^+$, 295 $(C_{18}H_{15}O_2S)^+$, 72 $(C_4H_{10}N)^+$; Anal. Calcd for $C_{22}H_{25}NO_2S$: C, 71.90; H, 6.86; N, 3.81. Found: C, 71.92; H, 6.83; N, 3.85.

4.1.1.28. Diethyl-(2-{4-[(2-methoxy-phenyl)-thiophen-2-yl-methyl]-phenoxy}-ethyl)-amine ***(30)****.* As described above, compound **4** *o*-OMe substituted at Ar^1 (400 mg, 1.349 mmol)refluxed with K_2CO_3 (466.32 mg, 3.37 mmol), 1-(2-chloroethyl)-diethyl amine hydrochloride (232.16 mg, 1.349 mmol) in acetone furnished **30** (360 mg, 67.5%) as brown viscous oil; ^1H NMR (300 MHz, $CDCl_3$): δ 7.24–7.02 (m, 5H), 6.91–6.79 (m, 5H), 6.63–6.62 (m, 1H), 6.03 (s, 1H), 4.02 (t, *J* = 6.3, 2H), 3.73 (s, 3H), 2.85 (t, *J* = 6.3, 2H), 2.66–2.59 (m, 4H), 1.05 (t, *J* = 7.1, 6H); ^{13}C NMR (300 MHz, $CDCl_3$): δ 157.3, 156.6, 148.6, 135.9, 132.8, 129.7 (2C), 129.5, 127.7, 126.3, 125.9, 123.9, 120.3, 114.1 (2C), 110.6, 66.3, 55.5, 51.7, 47.7 (2C), 43.8, 11.7 (2C); IR (Neat): 3017, 2971, 2935, 2836, 2362, 1607, 1509, 1244, 1217, 764 cm^{-1}; MS (ESI): *m/z* 396 $(M+H)^+$; Anal. Calcd for $C_{24}H_{29}NO_2S$: C, 72.87; H, 7.39; N, 3.54. Found: C, 72.95; H, 7.36; N, 3.58.

4.1.1.29. 1-(2-{4-[(2-Methoxy-phenyl)-thiophen-2-yl-methyl]-phenoxy}-ethyl)-pyrrolidine ***(31)****.* Compound **4** *o*-OMe substituted at Ar^1 (500 mg, 1.687 mmol) refluxed with K_2CO_3 (582.83 mg, 4.217 mmol), 1-(2-chloroethyl)-pyrrolidine hydrochloride (286.92 mg, 1.687 mmol) in acetone furnished **31** (478.6 mg, 72.1%) as brown viscous oil; ^1H NMR (300 MHz, $CDCl_3$): δ 7.24–6.98 (m, 5H), 6.88–6.77 (m, 5H), 6.60–6.59 (m, 1H), 5.99 (s, 1H), 4.09 (t, *J* = 5.9, 2H), 3.73 (s, 3H), 2.92 (t, *J* = 5.9, 2H), 2.68 (bs, 4H), 1.84–1.80 (m, 4H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 157.0, 156.5, 148.4, 136.1, 132.6, 129.7 (2C), 129.5, 127.7, 126.3, 125.9, 123.9, 120.3, 114.1 (2C), 110.4, 66.1, 55.4, 54.7, 54.5 (2C), 43.8, 23.4 (2C); IR (Neat): 3017, 2964, 2601, 1607, 1509, 1224, 1217, 769 cm^{-1}; MS (ESI): *m/z* 394 $(M+H)^+$; Anal. Calcd for $C_{24}H_{27}NO_2S$: C, 73.25; H, 6.92; N, 3.56. Found: C, 73.18; H, 6.95; N, 3.54.

4.1.1.30. 1-(2-{4-[(2-Methoxy-phenyl)-thiophen-2-yl-methyl]-phenoxy}-ethyl)-piperidine ***(32)****.* Compound **4** *o*-OMe substituted at Ar^1 (500 mg, 1.687 mmol) refluxed with K_2CO_3 (582.83 mg, 4.217 mmol), 1-(2-chloroethyl)-piperidine hydrochloride (310.59 mg, 1.687 mmol) in acetone furnished **32** (630 mg, 91.62%) as brown viscous oil; ^1H NMR (300 MHz, $CDCl_3$): δ 7.11–6.99 (m, 5H), 6.88–6.76 (m, 5H), 6.61–6.59 (m, 1H), 5.99 (s, 1H), 4.06 (t, *J* = 6.0, 2H), 3.73 (s, 3H), 2.76 (t, *J* = 5.9, 2H), 2.52 (t, *J* = 5.2, 4H), 1.61 (q, *J* = 5.6, 4H), 1.47–1.42 (m, 2H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 157.2, 156.6, 148.5, 136.0, 132.8, 129.8, 129.6, 127.7, 126.3, 126.0, 123.9, 120.4, 114.2 (2C), 110.5, 96.2 65.5, 57.8, 55.5, 54.9 (2C), 43.8, 25.6 (2C), 24.1; IR (Neat): 3020, 2962, 260, 1610, 1217, 1091, 929, 761 cm^{-1}; MS (ESI): *m/z* 408 $(M+H)^+$; Anal. Calcd for $C_{25}H_{29}NO_2S$: C, 73.67; H, 7.17; N, 3.44. Found: C, 73.76; H, 7.21; N, 3.34.

4.1.1.31. 1-(2-{4-[(2-Methoxy-phenyl)-thiophen-2-yl-methyl]-phenoxy}-ethyl)-azepane ***(33)****.* Compound **4** *o*-OMe substituted at Ar^1 (200 mg, 0.674 mmol) refluxed with K_2CO_3 (233.16 mg, 1.68 mmol), 1-(2-Chloro-ethyl)-azepane hydrochloride (265.47 mg, 0.674 mmol) in acetone furnished **33** (197 mg, 69.2%) as. brown viscous oil; ^1H NMR (300 MHz, $CDCl_3$): δ 7.19–6.98 (m, 5H), 6.87–6.75 (m, 5H), 6.60–6.58 (m, 1H), 5.99 (s, 1H), 4.04 (t, *J* = 5.8, 2H), 3.73 (s, 3H), 2.95 (t, *J* = 5.5, 2H), 2.79 (t, *J* = 4.2, 4H), 1.67 (bs, 4H), 1.60 (bs, 4H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 157.2, 156.6, 148.5, 136.1, 132.8, 129.8 (2C), 129.6, 127.7, 126.3, 125.9, 123.9, 120.4, 114.2 (2C), 110.5, 65.8, 56.5, 55.7, 55.4 (2C), 43.8, 27.3 (2C), 27.0 (2C); IR

(Neat): 3017, 2937, 1609, 1509, 1242, 1217, 767 cm^{-1}; MS (ESI): *m/z* 422 $(M+H)^+$; 126 $(C_8H_{16}N)^+$; Calcd for $C_{26}H_{31}NO_2S$: C, 74.07; H, 7.41; N, 3.32. Found: C, 74.02; H, 7.39; N, 3.33.

*4.1.1.32. Diisopropyl-(2-{4-[(2-methoxy-phenyl)-thiophen-2-yl-methyl]-phenoxy}-ethyl)-amine **(34)**.* Compound **4** *o*-OMe substituted at Ar^1 (500 mg, 1.687 mmol) refluxed with K_2CO_3 (582.83 mg, 4.217 mmol, (2-Chloro-ethyl)-diisopropyl-amine hydrochloride (337.65 mg, 1.687 mmol) in acetone furnished **34** (520 mg, 72.7%) as yellow viscous oil; ^1H NMR (300 MHz, $CDCl_3$): *δ* 7.17–6.98 (m, 5H), 6.88–6.78 (m, 5H), 6.60–6.59 (m, 1H), 5.99 (s, 1H), 3.87 (bs, 2H), 3.74 (s, 3H), 3.06 (bs, 2H), 2.82 (t, *J* = 6.7, 2H), 1.05 (d, *J* = 6.2, 12H); ^{13}C NMR (75 MHz, $CDCl_3$): *δ* 157.7, 156.8, 135.9, 133.0, 129.9 (2C), 129.7 (2C), 127.9, 126.5, 126.1, 124.1, 120.5, 114.2 (2C), 110.8, 69.3, 55.6, 49.8 (2C), 44.6, 44.0, 21.0 (4C); IR (Neat): 3018, 2968, 2930, 2361, 1605, 1508, 1217, 764 cm^{-1}; MS (ESI): *m/z* 424 $(M+H)^+$; Anal. $C_{26}H_{33}NO_2S$: C, 73.72; H, 7.85; N, 3.31. Found: C, 73.81; H, 7.90; N, 3.25.

*4.1.1.33. 4-(2-{4-[(2-Methoxy-phenyl)-thiophen-2-yl-methyl]-phenoxy}-ethyl)-morpholine **(35)**.* Compound **4** *o*-OMe substituted at Ar^1 (400 mg, 1.349 mmol), K_2CO_3 (466.32 mg, 3.37 mmol), 4-(2-Chloro-ethyl)-morpholine hydrochloride (251.02 g, 1.349 mmol) furnished **35** (510 mg, 92.2%) as brown viscous oil. ^1H NMR (200 MHz, $CDCl_3$): *δ* 7.23–6.98 (m, 5H), 6.88–6.75 (m, 5H), 6.60–6.59 (m, 1H), 5.99 (s, 1H), 4.05 (t, *J* = 5.7, 2H), 3.73 (s, 3H), 3.69 (t, *J* = 4.5, 4H), 2.75 (t, *J* = 5.7, 2H), 2.53 (t, *J* = 4.6, 4H); ^{13}C NMR (75 MHz, $CDCl_3$): *δ* 157.2, 156.6, 148.4, 136.1, 132.7, 129.8 (2C), 129.6, 127.8, 126.3, 126.0, 123.9, 120.4, 114.1 (2C), 110.5, 66.8 (2C), 65.6, 57.7, 55.4, 54.3 (2C), 43.8; IR (Neat): 3012, 2936, 2863, 1605, 1508, 1243, 1220, 767 cm^{-1}. MS (ESI): *m/z* 410 $(M+H)^+$; Anal. Calcd for $C_{24}H_{27}NO_3S$: C, 70.39; H, 6.65; N, 3.42. Found: C, 70.43; H, 6.72; N, 3.43.

*4.1.1.34. Diisopropyl-(2-{4-[(4-methoxy-phenyl)-thiophen-2-yl-methyl]-phenoxy}-ethyl)-amine **(36)**.* Compound **4** *p*-OMe substituted at Ar^1 (0.10 g, 0.33 mmol), K_2CO_3 (0.14 g, 1.01 mmol), (2-Chloro-ethyl)-diisopropyl-amine hydrochloride (0.07 g, 0.37 mmol) furnished **36** as yellow viscous oil (0.11 g, 75%). 1H NMR (300 MHz, CDCl3): *δ* 7.17 (d, *J* = 4.5, 1H), 7.12–7.07 (m, 4H), 6.92–6.89 (m, 1H), 6.84–6.80 (m, 4H), 6.67–6.64 (d, *J* = 3.0, 1H), 5.56 (s, 1H), 3.86 (t, *J* = 7.3, 2H), 3.76 (s, 3H), 3.07–2.98 (m, 2H), 2.80 (t, *J* = 7.5, 2H), 1.03 (d, *J* = 6.3, 12H); 13C NMR (75 MHz, CDCl3): *δ* 158.2, 157.5, 148.9, 136.3, 136.2, 129.6 (4C), 126.4, 125.9, 124.2, 114.2 (2C), 113.6 (2C), 68.8, 55.1 (2C), 50.5, 49.9, 44.5, 20.6 (4C); IR (Neat): 2964, 1654, 1611, 1509 cm^{-1}; MS (ESI): *m/z* 424 $(M+H)^+$; Anal. Calcd for $C_{26}H_{33}NO_2S$: C, 73.72; H, 7.85; N, 3.31. Found: C, 73.79; H, 7.79; N, 3.29.

*4.1.1.35. 1-(2-(4-((4-Methoxyphenyl) (thiophen-3-yl)methyl)phenoxy)ethyl)piperidine **(37)**.* Compound **1a** reacted with 2* to gave corresponding 3* which on refluxing with phenol and two drops of H_2SO_4 in benzene provided with 4* which was refluxed with K_2CO_3 1-(2-chloroethyl)-piperidine hydrochloride in acetone furnished compound **37**. ^1H NMR (400 MHz, $CDCl_3$): δ 7.00 (d, J = 8 Hz, 1H), 6.95 (m, 4H), 6.77–6.63 (m, 5H), 6.62 (s,1H), 7.32 (s,1H), 4.32 (t, J = 4 Hz, 2H), 3.7 (s, 3H), 3.27 (t, J = 4 Hz, 2H), 3.09 (m,4H), 1.88 (m,4H), 1.59 (m,2H); ^{13}C NMR (75 MHz, CDCl3): *δ* 158.11, 155.7, 145.3, 137.7, 136.0, 130.0 (2C), 129.7 (2C), 125.5 (2C), 122.4, 114.3 (2C), 113.7 (2C), 63.0, 56.3, 55.2 (2C), 54.1, 50.9, 23.0 (2C), 21.9.

*4.1.1.36. N, N –diethyl-2-(4-((4-methoxyphenyl) (thiophen-3-yl) methyl)phenoxy)ethanamine **(38)**.* R_f: 0.4 (20% ethyl acetate in hexane). Isolated as light yellow oily liquid (yield 65%) by elution with 2% methanol in dichloromethane from silica gel. ^1H NMR (400 MHz, $CDCl_3$): δ 7.19 (t, J = 4 Hz,1H), 7.0 (m, 4H), 6.97 (m, 5H), 6.76–6.62 (m, 1H), 5.32 (s, 1H), 4.29 (t, J = 4 Hz,2H), 3.70 (s, 3H), 3.43 (t, J = 4 Hz, 2H), 3.23 (m, 4H), 1.31 (t, J = 8 Hz, 6H).

*4.1.1.37. N-isopropyl-N-(2-(4-((4-methoxyphenyl) (thiophen-3-yl) methyl)phenoxy)ethyl)propan-2-amine **(39)**.* R_f: 0.43 (20% ethyl acetate in hexane). Isolated as light yellow oily liquid (yield 62%) by elution with 2% methanol in dichloromethane from silica gel. ^1H NMR (300 MHz, $CDCl_3$): δ 7.23 (s, 1H), 7.03 (m, 4H), 6.85 (m, 5H), 6.69 (m, 1H), 5.39 (s, 1H), 3.87 (t, J = 6 Hz, 2H), 3.76 (s, 3H), 3.03 (t, J = 6 Hz, 2H), 2.80 (m, 2H), 1.04 (d, J = 6 Hz, 12H).

*4.1.1.38. 2-(Di(thiophen-2-yl)methoxy)-N,N-diethylethanamine **(40)**.* To a solution of 2-thiophene carboxaldehyde, thiophene 2-magnesium bromide (*in situ* generation of 2-bromo thiophene in presence of Mg in THF for 1hr) was added and was stirred for another 1hr to furnish corresponding carbinol. It was then refluxed with diethyl aminoethyl chloride hydrochloride chain in presence of K_2CO_3 in acetone at 60–75 °C to furnish compound **40**. Quenched with NH_4Cl soln, extracted with ethyl acetate and dried over Na_2SO_4. The solvent was removed under reduced pressure. Product was purified by column chromatography. R_f: 0.3 (2% methanol in DCM). Isolated as pale yellow, oily liquid, (yield 52%), elution with 1.5% methanol in DCM from silica gel. ^1H NMR (400 MHz, $CDCl_3$): δ 7.23 (s, 2H), 6.90 (s, 4H), 5.83 (s, 1H), 3.72 (s, 2H), 3.00–2.90 (m, 6H), 1.16 (d, *J* = 6.48 Hz, 6H)ppm. ^{13}C NMR (75 MHz, $CDCl_3$): δ 145.1 (2C), 126.3 (2C), 125.7 (2C), 125.6 (2C), 75.9, 66.5, 51.8, 47.6 (2C), 11.1 (2C) ppm. IR (Neat cm^{-1}): 3442, 2924, 1630, 1218, 772. Anal. calcd. for $C_{13}H_{17}NOS_2$: C 58.39; H, 6.41; N, 5.24%; found C, 58.42; H, 6.34; N, 5.15%.

*4.1.1.39. N-(2-(dithiophen-2-ylmethoxy)ethyl)-N-isopropylpropan-2-amine **(41)**.* R_f: 0.6 (40% ethyl acetate in hexane). Isolated as pale brown, oily liquid, (yield 70%) by elution with 0.5% methanol in DCM from silica gel. ^1H NMR (300 MHz, $CDCl_3$): δ 7.21 (d, *J* = 4.89 Hz, 2H), 6.88 (t, *J* = 5.07 Hz,4H), 5.85 (s, 1H), 3.64 (s, 2H), 3.29 (s, 2H), 2.87 (s, 2H), 1.10 (d, *J* = 6.09 Hz, 12H)ppm. IR (Neat cm^{-1}):3440, 2928, 1642, 1218, 770. Anal. calcd. for $C_{17}H_{25}NOS_2$: C, 63.11; H, 7.79; N, 4.33%; found C, 63.24; H, 7.65; N, 4.41%.

*4.1.1.40. 2-(Di(thiophen-2-yl)methoxy)-N,N-dimethylethanamine **(42)**.* R_f: 0.35 (2% methanol in DCM). Isolated as pale brown, oily liquid, (yield 35%) by elution with 2% methanol in DCM from silica gel. ^1H NMR (300 MHz, $CDCl_3$): δ 7.25 (d, *J* = 4.5 Hz, 2H), 6.91 (d, *J* = 4.44 Hz, 4H), 5.85 (s, 1H), 3.87 (t, *J* = 3.69 Hz, 2H), 3.25 (d, *J* = 3.75 Hz, 2H), 2.85 (s, 6H) ppm. ^{13}C NMR (100 MHz, $CDCl_3$): δ 143.7 (2C), 126.6 (2C), 126.3 (2C), 126.1 (2C), 76.3, 63.8, 57.3, 44.3 (2C) ppm. IR (Neat cm^{-1}): 3435, 2924, 1634, 1217, 770. Anal. calcd. for $C_{15}H_{21}NOS_2$: C, 60.98; H, 7.16; N, 4.74%; found C, 60.77; H, 7.32; N, 4.71%.

*4.1.1.41. 1-(2-(Di(thiophen-2-yl)methoxy)ethyl)piperidine **(43)**.* R_f: 0.4 (5% methanol in DCM). Isolated as pale orange, oily liquid, (yield 33%) by elution with 1.5% methanol in DCM from silica gel. ^1H NMR (400 MHz, $CDCl_3$): δ 7.28(dd, J_1 = 1.68, J_2 = 2.88 Hz, 2H), 6.88 (t, *J* = 4.64 Hz, 4H), 5.82 (s, 1H), 3.65 (t, *J* = 5.88 Hz, 2H), 2.66 (t, *J* = 5.84 Hz, 2H), 2.51 (s, 4H), 1.61–1.55 (m, 4H),1.39 (d, *J* = 4.96 Hz, 2H)ppm. IR (Neat cm^{-1}):3450, 2918, 1620, 1212, 775. ($C_7H_{14}NO$). Anal. calcd. for $C_{16}H_{21}NOS_2$: C, 62.50; H, 6.88; N, 4.56%; found C, 62.63; H, 6.78; N, 4.56%.

*4.1.1.42. N-(2-(di(thiophen-3-yl)methoxy)ethyl)-N-isopropylpropan-2-amine **(44)**.* R_f: 0.38 (20% ethyl acetate in hexane). Isolated as yellow oily liquid (yield 64%) by elution with 2% methanol in dichloromethane from silica gel. ^1H NMR (400 MHz, $CDCl_3$):

δ 7.20–7.17 (m, 4H), 7.01 (q, *J* = 1 Hz,1H), 6.83 (d, J = 5.28 Hz, 1H), 5.7 (s, 1H), 3.5 (s, 2H), 3.05 (s, 2H), 2.70 (s, 2H), 0.99 (d, *J* = 5.84 Hz, 12H).

4.1.1.43. 1-(2-(Di(thiophen-3-yl)methoxy)ethyl)piperidine ***(45)***. R_f: 0.20 (30% ethyl acetate in hexane). Isolated as yellow oily liquid (yield 67%) by elution with 5% methanol in dichloromethane from silica gel. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.22–7.19 (m, 4H), 6.99 (q, *J* = 1.6 Hz,1H), 6.86 (d, J = 5.28 Hz, 1H), 5.76 (s, 1H), 3.75 (t, J = 5.32 Hz, 2H), 2.39 (t, *J* = 5.36 Hz, 2H), 2.78 (s, 4H),1.75–1.69 (m, 4H), 1.52 (s, 2H).

4.1.1.44. 2-(Di(thiophen-3-yl)methoxy)-N,N-diethylethanamine ***(46)***. R_f: 0.30 (20% ethyl acetate in hexane). Isolated as yellow oily liquid (yield 68%) by elution with 6% methanol in dichloromethane from silica gel. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.18 (t, J = 4.56 Hz, 4H), 6.99 (q, *J* = 1.24 Hz, 1H), 6.83 (d, J = 5.28 Hz, 1H), 5.76 (s, 1H), 3.57 (t,J = 6.16 Hz, 2H), 2.72 (t, *J* = 6.2 Hz, 2H), 2.57 (q, J = 7.16 Hz, 4H),0.99 (t, J = 7.16 Hz, 6H).

4.1.1.45. 2-(Di(thiophen-3-yl)methoxy)-N,N-dimethylethanamine ***(47)***. R_f: 0.30 (20% ethyl acetate in hexane). Isolated as Brown yellow oily liquid (yield 72%) by elution with 10% methanol in dichloromethane from silica gel. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.20 (t, *J* = 3.24 Hz, 4H), 7.00 (d, *J* = 4.72 Hz, 1H), 6.852 (d, J = 5.28 Hz, 1H), 5.75 (s, 1H), 3.63 (t, J = 5.52 Hz, 2H), 2.69 (t, *J* = 5.52 Hz, 2H), 2.35 (s, 6H).

4.1.1.46. 1-(2-(Di(thiophen-3-yl)methoxy)ethyl)pyrrolidine ***(48)***. R_f: 0.30 (30% ethyl acetate in hexane). Isolated as pale yellow oily liquid (yield 68%) by elution with 4% methanol in dichloromethane from silica gel. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.25–7.20 (m, 4H), 7.00 (q, *J* = 1.48 Hz,1H), 6.86 (d, J = 5.28 Hz, 1H), 5.77 (s, 1H), 3.82 (q, J = 1.92 Hz, 2H), 3.14 (t, *J* = 4.28 Hz, 6H), 1.95 (t, J = 6.6 Hz, 4H).

4.1.1.47. 4-(2-(Di(thiophen-3-yl)methoxy)ethyl)morpholine ***(49)***. R_f: 0.36 (10% ethyl acetate in hexane). Isolated as yellow oily liquid (yield 67%) by elution with 2% methanol in dichloromethane from silica gel. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.20 (q, J = 1.52 Hz, 4H), 6.99 (q, *J* = 1.72 Hz, 1H), 6.86 (d, J = 5.28 Hz,1H), 5.76 (s,1H), 3.72 (t, J = 4.64 Hz, 4H), 3.68 (t, J = 5.48 Hz, 2H), 2.74 (t, *J* = 5.36 Hz, 2H), 2.64 (s, 4H).

4.2. Biological evaluation

4.2.1. Assay for in vitro activity against M. tuberculosis H37R$_V$ [11]

The compounds were dissolved in dimethyl sulfoxide (DMSO) to make stocks. Serial dilutions from stocks were also made in DMSO. To 1.9 mL Middle brook (MB) 7H10 agar supplemented with OADC (in tubes, held at 45–50 °C), 0.1 mL of test compound or DMSO (negative control) or INH (positive control) was added. The contents were mixed and allowed to solidify (by cooling) as slants. Three-week old culture of *M. tuberculosis* H37R$_v$ was harvested from L-J medium and its suspension (1 mg/mL, equivalent to 10^8 bacilli approximately) was made in normal saline containing 0.05% Tween-80. 10 μL of 1:10 dilution of this suspension (~10^5 bacilli) was inoculated into each tube and incubated at 37 °C for 4 weeks. The lowest concentration of a compound up to which there was no visible growth of bacilli was its minimal inhibitory concentration (MIC). Compounds having MIC of ≤6.25 mg/L were selected as 'secondary hits' for further development.

4.2.2. Assays for in vitro cytotoxicity [12]

In vitro cytotoxicity of active compounds (having MIC ≤ 6.25 mg/L) was measured using Vero cells and mouse bone-marrow derived macrophages. Vero C-1008 cell line was procured from Laboratory Animals Division (LAD) of CDRI. For preparation of macrophages, a Swiss mouse (bred in LAD) was euthanized by exposure to CO_2 and femurs dissected out. The bones were trimmed at each end and marrow flushed out with Dulbecco's Minimal Essential Medium containing 10% foetal bovine serum (DMEM-FBS) and antibiotics. The medium was also supplemented with 15% (*v*/*v*) L929 fibroblast conditioned medium and non-essential amino acids. The cell suspension (Vero/macrophage) was plated in 96-well tissue culture plates (20,000 cells/200 μL/well) and incubated overnight (37 °C, 5% CO_2) to allow their adherence. Compounds at different concentrations were added to the wells. A known toxic compound (Staurosporine) was used as a positive control and DMSO was used as negative control. After 24 h of incubation, 20 μL of MTS solution (tetrazolium compound, Owen's reagent) was added to each well and incubated for further 2 h (37 °C, 5% CO_2). O. D. was read at 490 nm using a plate reader. A compound was considered as potentially toxic if its IC_{50} (concentration causing 50% loss in cell viability) was ≤10 times its MIC for *M. tuberculosis* H37R$_V$.

4.2.3. Assay for ex vivo activity in the macrophage model of tuberculosis [13]

The effect of selected compounds (MIC ≤ 6.25 mg/L and SI ≥ 10, i.e., non-toxic) on the survival and multiplication of *M. tuberculosis* H37R$_v$ within mouse bone-marrow derived as well as human blood monocyte derived macrophages was evaluated by light microscopy and counting of colony-forming units (CFU). Mouse bone-marrow derived macrophages were prepared as above and the human blood monocyte derived macrophages were prepared as follows: 10 mL blood from healthy volunteers was collected in anti-coagulant solution (acid-citrate-dextrose, ACD), centrifuged at 2500 rpm (to remove plasma) and reconstituted with RPMI 1640 tissue culture medium containing ACD. This suspension was layered over Ficoll-Isopaque (Sigma Chemical Company, USA) and centrifuged at 600 g for 20 min. The mononuclear cells on the top of Ficoll layer were collected, washed twice with RPMI medium, and counted using hemocytometer. The cells were finally suspended at a density of 10^6 cells/mL in RPMI containing heat inactivated pooled normal human serum (PNS, 5%).

For evaluation by microscopy, macrophages (human/mouse) were seeded in 8-chambered slides at a density of 5×10^5 cells (in 0.5 mL RPMI-PNS/DMEM-FBS media) per chamber. The cells were incubated at 37 °C with 5% CO_2. On 6th day, washed adherent macrophages in each chamber were infected for 3 h with 0.5 mL suspension containing 2.5×10^6 *M. tuberculosis* H37R$_v$ in antibiotic-free medium. Later, the chambers were thoroughly washed to remove extracellular bacteria and replenished with fresh 0.5 mL medium containing 4× MIC of standard drugs (INH, RFM and PZA) or test compounds. After further 4 days incubation in the CO_2 incubator, each chamber was gently washed and medium aspirated off. After removing the chambers, infected macrophages on slide were heat-fixed, stained with Ziehl-Neelsen stain, counter stained with methylene blue, and viewed under oil immersion lens (100×) of a microscope.

For evaluation by counting colony forming units (CFU), 5-day old cultures of adherent macrophages (10^6 cells/well, in 24-well plates) were infected for 3 h with 5×10^6 *M. tuberculosis* H37R$_v$ in antibiotic-free medium. Later, the wells were washed to remove extracellular bacteria and replenished with fresh 1 mL antibiotic-free medium containing 4× MIC of standard drugs or test compounds. In order to determine the number of bacilli phagocytosed during the 3 h infection period (0 day count), one well was lysed with 0.1% saponin (15 min) and lysate (50 μL of 1:100 dilution) plated on MB 7H11 agar (in petri dishes) for colony counting. Other

wells, after further 4 days of incubation, were gently washed, cells lysed and lysates plated on MB 7H11 agar plates. CFUs were counted after 4 weeks of incubation at 37 °C.

4.2.4. Assay for in vivo activity in the mouse model of tuberculosis [14]

Outbred Swiss mice (obtained from CDRI-LAD) were infected with *M. tuberculosis* $H37R_v$ (10^7 CFU/mouse, i.v.) and divided into groups of 8–10 animals. Each experimental group received daily oral dose of the test compound (100 mg/kg body weight, for 28 days) dissolved in a suitable vehicle (water/corn oil, depending on its solubility profile). The drug-treated control groups received INH (25 mg/kg) or EMB (10 mg/kg) or PZA (150 mg/kg body weight) for 28 days. The untreated control groups received only the vehicle. At least 3 mice from each group were sacrificed on day 30 for determination of viable bacilli in the lungs. Serial dilutions of lung homogenates were placed on MB 7H11 agar medium and colonies (CFU) were counted after 4 weeks of incubation at 37 °C. Remaining mice, if survived, were observed for further 15 days for determination of mean survival time (MST) and % survivors.

4.2.5. Determination of minimum bactericidal concentration (MBC) [15]

For *in vitro* MBC determinations, to each culture tube having 2.5 mL MB7H9 broth, 50 μL inoculum containing 10^5*M. tuberculosis* $H37R_v$ was added and incubated (37 °C) with or without 1× and 4× MIC of test compounds or standard *anti*-TB drugs. CFU in a tube containing inoculum alone was determined on day 0 and in others it was determined on day 14. The same principle was applied to the determination of *ex vivo* MBC.

4.2.6. Activity of selected compounds against drug-resistant strains of M. tuberculosis [11]

The drug sensitive and drug-resistant Indian clinical isolates of *M. tuberculosis* were obtained from the DBT Mycobacterium Repository at NJIL-OMD, Agra (Courtesy of Dr VM Katoch, DG-ICMR) and propagated on L-J medium. The cultures thus obtained were used for testing *in vitro* sensitivity against selected active compounds using agar dilution method.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.ejmech.2015.03.036.

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