

ONLINE SUPPORTING INFORMATION

Exploitation of a Novel Binding Pocket in Human Lipoprotein-Associated Phospholipase A2 (Lp-PLA₂) Discovered Through X-Ray Fragment Screening

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1. Computational Methods and Programs Used

All visualisation of the protein-ligand crystal structures was by AstexViewer.

<http://openastexviewer.net/web/overview.html>

Docking used the program GOLD. Jones, G.; Willett, P.; Glen, R. C.; Leach, A. R.; Taylor, R. Development and validation of a genetic algorithm for flexible docking. *J. Mol. Biol.* **1997**, 267, 727-748.

clogP calculation used the clogP reference manual. Daylight version 4.9.
<http://www.daylight.com/dayhtml/doc/clogp/> 2011.

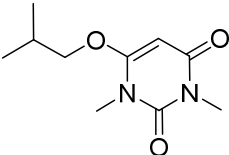
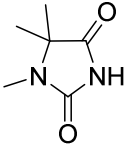
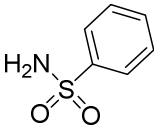
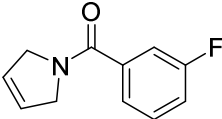
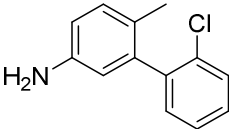
Molecular weight (in Daltons) was calculated by ChemDrawPro 14.0.

2. Recombinant Human Lp-PLA₂ PED6 Assay (IC₅₀ Determination)

N-((6-(2,4-Dinitrophenyl)amino)hexanoyl)-2-(4,4-difluoro-5,7-dimethyl-4-bora-3a,4a-diazas-indacene-3-pentanoyl)-1-hexadecanoyl-sn-glycero-3-phosphoethanolamine (PED6) is commercially available from Life Technologies. PED6 is an internally quenched fluorogenic substrate for Lp-PLA₂. Upon cleavage of the fluorogenic *sn*-2 acyl chain by Lp-PLA₂, the intramolecular quenching effect of the dinitrophenyl quencher on the lipid head group is relieved, resulting in a fluorescence increase.

The PED6 assay was used for compounds with weak activity against Lp-PLA₂ (IC₅₀ >10 μ M). Compound and 0.2 nM recombinant human Lp-PLA₂ enzyme in assay buffer (50 mM Tris pH 7.4, 150 mM NaCl, 1mM CHAPS) were incubated at room temperature for 30 minutes. 20 μ L PED6 substrate (6.25 μ M in assay buffer) was then added to each well and fluorescence (ex: 480 nm/ em: 540 nm) monitored continuously for 20 minutes using the Gemini plate reader (Molecular Devices). Initial reaction rates were calculated using SoftMax Pro, and IC₅₀ curves were generated in GraphPad Prism version 3 (La Jolla, CA, USA) and fitted using the four parameter logistic model. Individual assay results are reported in Supplementary Table 1.

Supplementary Table 1. Recombinant Human Lp-PLA₂ PED6 Assay Data

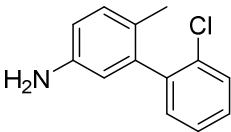
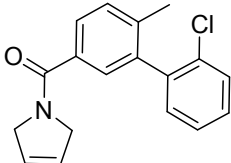
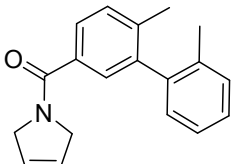
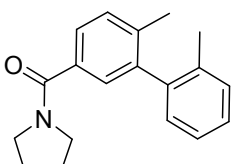
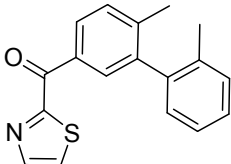
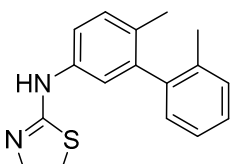
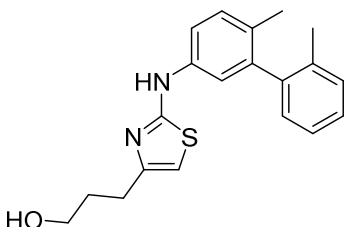
Compound	Structure	%I
2		66% at 1000 μ M 68% at 1000 μ M 71% at 1000 μ M
3		30% at 1000 μ M
4		20% at 1000 μ M
5		45% at 300 μ M 61% at 1000 μ M
6		49% at 100 μ M 50% at 100 μ M

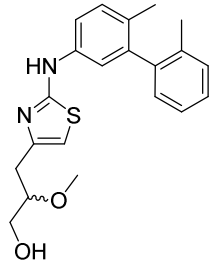
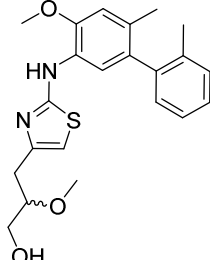
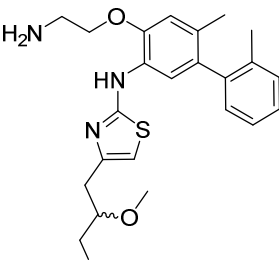
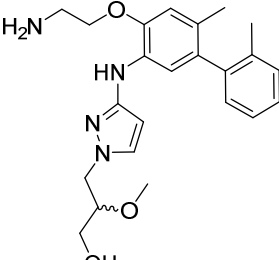
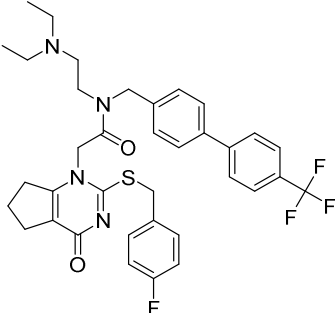
3. Recombinant Human Lp-PLA₂ Thio-PAF Assay (IC₅₀ Determination)

1-*O*-Hexadecyl-2-deoxy-2-thio-S-acetyl-sn-glycerol-3-phosphorylcholine (2-thio-PAF) is a substrate for PAF-hydrolases (PAF-AH) and is commercially available from Cayman Chemical. Upon cleavage with PAF-AH, the free thiol is released at the *sn*-2 position which can then react with the thiol-reactive coumarin 7-diethylamino-3-(4'-maleimidylphenyl)-4-methylcoumarin (CPM). This reaction (Michael addition) results in an increase in fluorescence. Inhibitors of Lp-PLA₂ prevent this cleavage and no increase in fluorescence is observed.

Compounds were incubated with 10 pM recombinant human Lp-PLA₂, followed by a substrate mix comprising 10 μM 2-Thio-PAF, 10 μM CPM and 100 μM *N*-ethylmaleimide in assay buffer (50 mM HEPES, pH 7.4, 150 mM NaCl, 1 mM CHAPS), in a final volume of 20 μL and incubated for 20 minutes at 25 °C. The plates were read for fluorescence intensity at ex: 380 nm / em: 485 nm using an Envision plate reader (Perkin Elmer). IC₅₀ curves were generated using the XL fit module in Microsoft Excel and fitted using 4 parameter logistic model or sigmoidal dose-response model. Individual assay results are reported in Supplementary Table 2, which indicates the reproducibility of the assay.

Supplementary Table 2. Recombinant Human Lp-PLA₂ Thio-PAF Assay Data

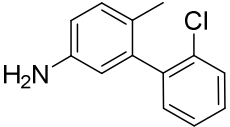
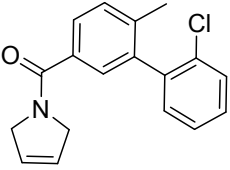
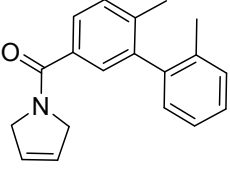
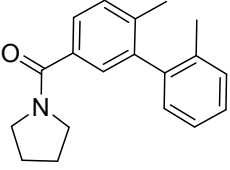
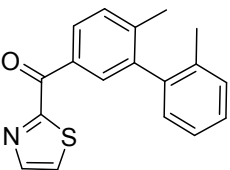
Compound	Structure	%I or IC ₅₀ (μM)	Geometric Mean IC ₅₀ (μM)
6		65% at 100 μM	~100
		40% at 100 μM	
7		0.12	0.18
		0.18	
		0.26	
8		0.15	0.23
		0.16	
		0.51	
9		15	30
		22	
		29	
		34	
		46	
		55	
10		1.9	2.0
		2.2	
11		1.6	1.7
		1.8	
		1.9	
12		0.23	0.23
		0.24	

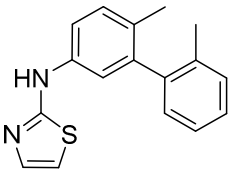
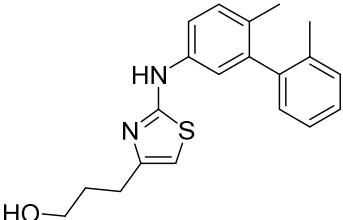
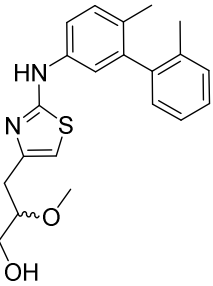
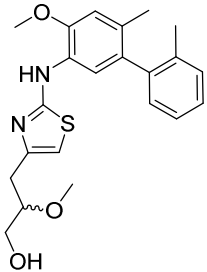
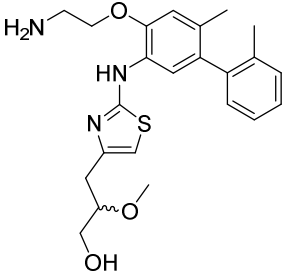
Compound	Structure	%I or IC ₅₀ (μM)	Geometric Mean IC ₅₀ (μM)
(±)-13		0.0071	0.0095
		0.013	
(±)-14		0.0032	0.0036
		0.0041	
(±)-15		0.0014	0.0024
		0.0026	
		0.0028	
		0.0037	
(±)-16		0.00091	0.0014
		0.0020	
Darapladib (1)		0.000026	0.000049
		0.000043	
		0.00011	

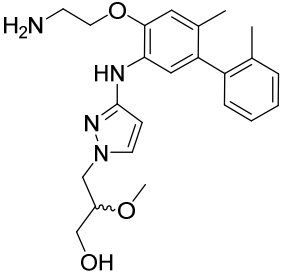
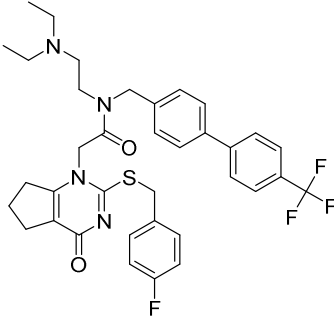
4. Recombinant Human PLA2-VIIB Thio-PAF Assay (IC₅₀ Determination)

Follow the procedure for recombinant human Lp-PLA₂ Thio-PAF assay (IC₅₀ determination), except using 100 pM of the recombinant human PLA2-VIIB enzyme instead of 10 pM recombinant human Lp-PLA₂. Individual assay results are reported in Supplementary Table 3, which indicates the reproducibility of the assay.

Supplementary Table 3. Recombinant Human PLA2-VIIB Thio-PAF Assay Data

Compound	Structure	IC ₅₀ (μM)	Geometric Mean IC ₅₀ (μM)
6		36	43
		53	
7		>10	>10
		>10	
		>10	
8		83	89
		90	
		96	
9		>100	>100
		>100	
		>100	
10		2.6	3.2
		3.9	

Compound	Structure	IC ₅₀ (μM)	Geometric Mean IC ₅₀ (μM)
11		6.1	11
		13	
		16	
12		>10	>10
		>10	
(±)-13		>10	>10
		>10	
(±)-14		6.1	6.1
		>10	
(±)-15		13	>10
		>10	
		>10	

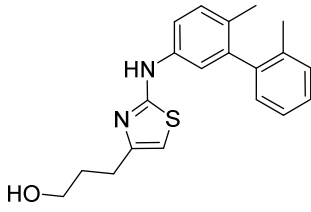
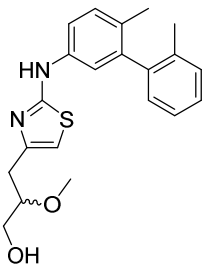
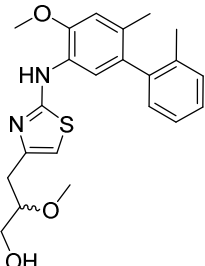
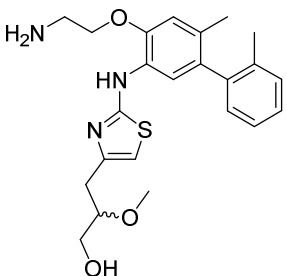
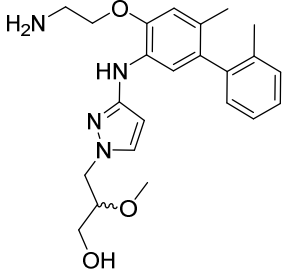
Compound	Structure	IC ₅₀ (μM)	Geometric Mean IC ₅₀ (μM)
(±)-16		>1	>1
		16	
Darapladib (1)		0.021	0.063
		0.047	
		0.062	
		0.094	
		0.095	
		0.12	

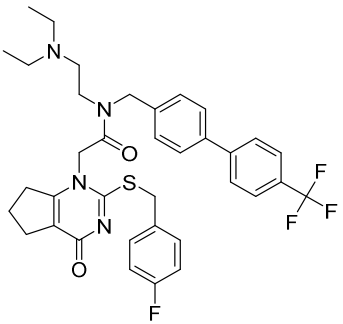
5. Lp-PLA₂ Human Plasma Thio-PAF Assay (IC₅₀ Determination)

The human plasma assay utilizes the same thioester analogue of PAF as described in the recombinant human Lp-PLA₂ Thio-PAF assay. This assay detects the activity of Lp-PLA₂ in human plasma, as determined by specific inhibition by Lp-PLA₂ inhibitors.

A 5 μ L aliquot of compound was added to 10 μ L pooled human plasma and the 96 well microplates were centrifuged for 30 seconds at 1000 rpm. After a 15 minute preincubation at room temperature, 5 μ L of substrate solution comprising 2 mM 2-thio-PAF [from ethanol stock], 52 μ M CPM [from a DMSO stock] and 2.5 mM NEM [made fresh daily in DMSO] in assay buffer (50 mM HEPES, pH 7.4, 150 mM NaCl, 1 mM CHAPS) was added. After 3 minutes, the reaction was quenched with 10 μ L of 5% trifluoroacetic acid. Plates were then centrifuged 30 seconds at 1000 rpm, covered to protect from light, and incubated for 10 minutes at room temperature. Plates were read at ex 380 nm, em 485 nm using an Envision plate reader (Perkin Elmer). IC₅₀ curves were generated using the XL fit module in Microsoft Excel. Individual assay results are reported in Supplementary Table 4, which indicates the reproducibility of the assay.

Supplementary Table 4. Lp-PLA₂ Human Plasma Thio-PAF Assay Data

Compound	Structure	IC ₅₀ (μM)	Geometric Mean IC ₅₀ (μM)
12		5.0	7.1
		10	
(±)-13		0.50	0.56
		0.63	
(±)-14		0.32	0.32
		0.32	
(±)-15		0.025	0.028
		0.032	
(±)-16		0.016	0.018
		0.020	

Compound	Structure	IC ₅₀ (μM)	Geometric Mean IC ₅₀ (μM)
Darapladib (1)		0.0022	0.035
		0.0031	
		0.0033	
		0.0065	

6. Lp-PLA₂ Protein Crystallography

Protein Purification

Lp-PLA₂ (residues 46-428 with a C-terminal His6 tag) was expressed in *E. coli BL21 (DE3)* at 30 °C. Cell pellets from a 10 L expression were resuspended in 400 mL lysis buffer (50 mM Tris-HCl pH 7.8, 0.5 M NaCl, 1.5 mM β -ME, 10 mM CHAPS) and lysed by sonication on ice. Following clarification at 14 K rpm for 30 mins, the supernatant was tumbled with 5-10 mL NiNTA resin for 2 hours at 4 °C. The resin was washed extensively with lysis buffer prior to elution in 50 mL lysis buffer containing 200 mM imidazole. The eluted protein was concentrated to 10 mL, diluted to 50 mL in 20 mM Tris-HCl pH 7.8, 5 mM DTT, 10 mM CHAPS and loaded onto a HitrapQ FF column and elute with a 0-1 M NaCl gradient. Peak fractions were concentrated to 10 mL and loaded onto a Hi Load Sephacryl 200 26/60 column equilibrated in 20 mM Tris-HCl pH 7.8, 300 mM NaCl, 5 mM DTT, 10 mM CHAPS. Peak fractions were concentrated 10 mL, diluted to 50 mL with 20 mM Tris pH 7.8, 5 mM DTT, 0.04% Pluronic F68, 0.002% Tween80, loaded onto HitrapQ FF column and eluted with 0-500 mM NaCl gradient. Peak fractions were concentrated to 10-12 mg/mL and stored at -80 °C.

Crystallization

β -D-Octyl glucoside (β -OG) (37 mM final) was added to the protein immediately prior to setting up trays. Hanging drops (2 μ L protein + 1 μ L 1-1.3 M NaCl, 28-32% PEG3350, 0.1 M HEPES pH 7.4) were incubated at 20 °C and crystals appeared overnight and continued to grow for several days. To obtain protein complex structures crystals were soaked in 30-33% PEG3350, 0.1 M HEPES pH 7.4, 0.2 M NaCl, 0.04% pluronic F68, 37 mM β -OG (or 10 mM CHAPS) and compound dissolved in DMSO.

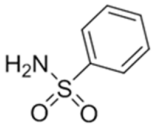
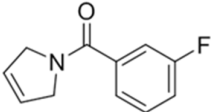
7. Protein Purification for Isothermal Titration Calorimetry (ITC), Thermal shift (T_m) and NMR

ITC, Thermal shift and NMR experiments were performed using an Lp-PLA₂ protein construct (residues 47-429 with an N-terminal His6 tag and TEV cleavage site) expressed in sf21 insect cells at 27 °C. Cells were harvested 72 hours after infection. Cell pellets were resuspended in lysis buffer (50 mM TrisHCl pH 7.8, 500 mM NaCl, 10 mM CHAPS and 1.5 mM β-mercaptoethanol), sonicated and clarified by centrifugation. The supernatant was then batch bound to NiNTA resin overnight at 4 °C. After extensively washing the resin in lysis buffer the bound protein was eluted in lysis buffer containing 250 mM imidazole. TEV protease was added to the eluted protein and the protein was then dialysed overnight at 4 °C in 20 mM TrisHCl pH 7.8, 10 mM CHAPS, 7.5% (w/v) sucrose and 2 mM β-mercaptoethanol. Following TEV cleavage the protein was loaded onto a Blue sepharose column (3 x 5 mL columns) equilibrated in 20 mM TrisHCl pH 7.8, 10 mM CHAPS, 7.5% (w/v) sucrose and 5 mM DTT. Protein was eluted with a 0-2 M NaCl gradient and fractions containing Lp-PLA₂ were pooled and then dialysed in 20 mM TrisHCl pH 7.8, 0.1% (w/v) Pluronic F-68, 7.5% (w/v) sucrose and 5 mM DTT. After 4 hours at 4 °C the dialysed protein was concentrated and then diluted ~20-fold into the same dialysis buffer. The protein was then captured on a HitrapQ FF column equilibrated in 20 mM TrisHCl pH 7.8, 0.1% (w/v) Pluronic F-68, 7.5% (w/v) sucrose and 5 mM DTT and eluted with a 0 - 500 mM NaCl gradient. Lp-PLA₂ containing fractions were pooled and concentrated before dialysis into a final buffer comprising 15 mM sodium citrate pH 6.5, 0.1% (w/v) Pluronic F-68 and 7.5% (w/v) sucrose. The protein was then aliquoted and stored at -80 °C.

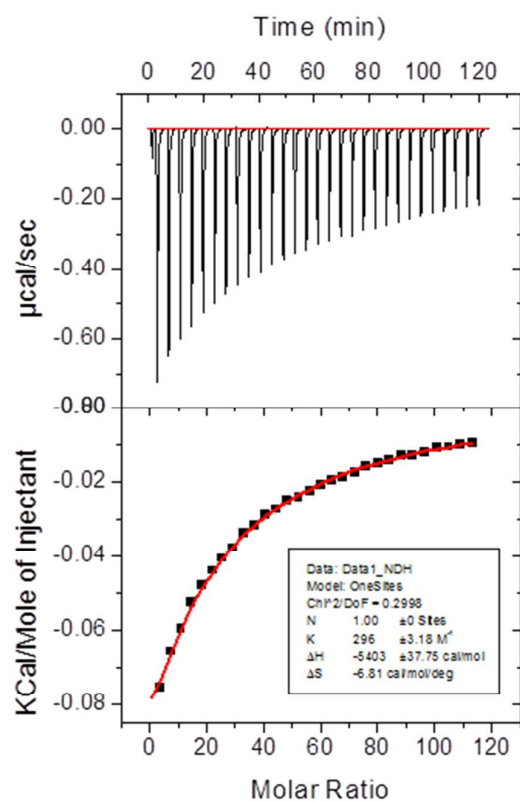
8. Lp-PLA₂ ITC Assay

ITC experiments were performed on a Microcal VP-ITC at 25 °C in a pH 7.5 buffer comprising 50 mM Hepes, 100 mM NaCl, 1 mM TCEP, 0.1% (w/v) Pluronic F68 and 5% DMSO. The Lp-PLA₂ protein used for the ITC experiments was prepared as described in Section 6. All ITC experiments were set up with protein in the sample cell and compound in the injection syringe. Data were fitted to a single site binding model using Origin 7.0 software. Data is reported in Supplementary Table 5 and Supplementary Figure 1.

Supplementary Table 5. Lp-PLA₂ Isothermal Titration Calorimetry (ITC) Assay

Compound	Structure	K _d (μM)	Replications
4		3400	n = 1
5		980	n = 1

Supplementary Figure 1: Fragment **4** binding to Lp-PLA₂ monitored by ITC.



9. Lp-PLA₂ T_m Assay

Protein purification is detailed within Section 6. Thermal shift screening was performed on a Stratagene MX3005p real-time PCR machine. Fluorescent dyes used for thermal shift assays typically exhibit a strong fluorescence signal in the presence of detergents. Since purified Lp-PLA₂ contains detergent (0.1% (w/v) Pluronic F68) it was necessary to dilute the protein into a detergent-free buffer. Therefore, for T_m shift experiments, Lp-PLA₂ was diluted ~1000-fold to a final concentration of 0.3 μM into a buffer comprising 50 mM Hepes, 100 mM NaCl and 2.5x Sypro orange at pH 7.5. Screening of the core fragment set (1360 fragments) gave 71 compounds with a ΔT_m of >2°C, corresponding to a hit rate of 5%.

10. Ligand Observed NMR Screening

Protein purification is detailed within Section 6. NMR fragment screening data were obtained for core fragment set (1360 fragments) using cocktails of 4 fragments and solutions containing 150 μL of 8 μM Lp-PLA₂ in 25 mM TRIS buffer pH 7.5, 150 mM NaCl, 5 mM DTT and 0.1% Pluronic F-68 detergent. The final solutions contained 500 μM of each fragment and 2% DMSO. Hits were identified using standard ligand-detected NMR methods. Control spectra, obtained from samples which contained fragments but no Lp-PLA₂, were subtracted from each dataset in order to remove any NMR signals that arose from weak binding to the detergent. In a second step, 60 μM of a tool compound was added to each protein-containing solution and the NMR cocktail screening experiment was repeated in a competition format. The tool compound binds to Lp-PLA₂ in the lipase active site with a K_d

of 230 nM (unpublished data) and should displace any weakly-bound fragments from this site. Subtraction of this second spectrum from the results obtained in the absence of the tool compound and in buffer alone allowed the total NMR signal obtained for each fragment to be divided into contributions from competitive and non-competitive binding to Lp-PLA₂, as well as binding to the detergent.

From 1360 fragments screened, 191 (14%) showed clear binding to one or more sites on Lp-PLA₂. Of these fragments, 63 (5%) showed significant competition with the tool compound. The remaining 128 hits bound non-competitively with the tool compound, possibly at multiple, non-specific sites.

11. In Vitro and PK Assays

11a. ChromLogD

The Chromatographic Hydrophobicity Index (CHI) values were measured using reversed phase HPLC column (Luna C18 (2), Phenomenex, UK) with a fast acetonitrile gradient at starting mobile phase of pH=7.4. CHI values are derived directly from the gradient retention times by using a calibration line obtained for standard compounds. The CHI value approximates to the volume % organic concentration when the compound elutes. CHI is linearly transformed into ChromlogD by least-square fitting of experimental CHI values to calculated clogP values for over 20,000 research compounds using the following formula: $\text{ChromlogD} = 0.0857\text{CHI} - 2.00$. The average error of the assay is ± 3 CHI unit or ± 0.25 ChromlogD.

11b. Chemi-Luminescent Nitrogen Detection (CLND) Solubility Assay

5 μL of 10 mM DMSO stock solution was diluted to 100 μL with pH 7.4 phosphate buffered saline, equilibrated for 1 hour at room temperature, filtered through Millipore Multiscreen_{HTS}-PCF filter plates (MSSL BPC). The filtrate was quantified by suitably calibrated flow injection CLND. The standard error of the CLND solubility determination is ± 30 μM , the upper limit of the solubility is 500 μM when working from 10 mM DMSO stock solution. For further details on CLND measurement, see *Tetrahedron Lett.* **1997**, 38, 1689-1692.

11c. Artificial Membrane Permeability Assay

GSK in-house artificial membrane permeability assay: A 1.8% lipid (phosphatidyl choline, egg) in 1% cholesterol decane solution was applied to a Millicell 96-well, 0.4 μm , PCF culture plate. 250 μL and 100 μL 50 mM phosphate buffer pH 7.4 with 0.5% encapsin was applied to the donor and receiver compartments, respectively. 2.5 μL of a 10 mM stock solution of compound in DMSO was added to the donor compartment. The assay was incubated at RT for 3 hours. Samples from both donor and receiver compartments were analyzed by HPLC with UV detection at 215 and 254 nm and permeability was calculated. The permeability ($\log P_{\text{app}}$) measures how fast molecules pass through the lipid membrane is expressed in nm/s.

11d. Human Plasma Protein Binding (hPPB) Assay

Chemically bonded Human Serum Albumin (HSA) and Alpha-1-acidglycoprotein HPLC stationary phases (Chiral Technologies, France) were used for measuring compounds' binding to plasma proteins, applying linear gradient elution up to 30% iso-propanol. The run time was 6 min, including the re-equilibration of the stationary phases with the 50 mM pH 7.4 ammonium acetate buffer. The obtained gradient retention times were standardised using a calibration set of mixtures as described in the reference *J. Pharm. Sci.* **2003**, 92, 2236–2248. The average standard error of the assay depends on the binding strength and kinetics of the compounds. It ranges from $\pm 5\%$ in the medium binding range which reduces to 0.1% at binding above 99% with fast kinetics.

11e. Microsomal Rat Clint Data

Microsomal protocol

The assay was designed for determining the stability of test compounds in microsomal matrix. Test compounds were dissolved in DMSO at 1 mg/mL. Daughter solution was prepared in methanol/water (50/50) at 30 µg/mL. Microsomes (rat liver) (from Xenothech) were diluted at 0.625 mg proteins per mL in phosphate buffer 50 mM pH 7.4. NADPH cofactor was prepared with glucose-6-phosphate (7.8 mg), glucose-6-phosphate dehydrogenase (6 units), NADP (1.7 mg) per 1 mL in NaHCO₃ 2%.

Pre-incubation consisted of mixing 5 µL of daughter solution with 395 µL diluted microsomes and warming it at 37 °C for 7 minutes. Incubations was started with the addition of 100 µL of NADPH. 50 µL of mixture were taken at 0, 3, 6, 12 and 30 minutes and crashed with 150 µL acetonitrile containing an internal standard. A control compound was included in parallel and tested with each species.

After 10 minutes, centrifugation at 4000 rpm, 2 µL of samples were injected on a liquid chromatography system and eluted with an Ascentis C18 column (50 × 2.1 mm id, 2.7 µm) with 0.1% formic acid in water (A) and 0.1% formic acid in acetonitrile (B), using the following elution 2 minute gradient: 5 to 95% B over 1.2 min, 95% B over 0.6 min and 0.1 min to re-equilibrate the column, at 0.5 mL/min at 50°C. Samples were analysed by mass spectrometry with an electrospray source, in positive mode with the following mass transitions:

(±)-15: 428.4>353.3

(±)-16: 411.4>368.3

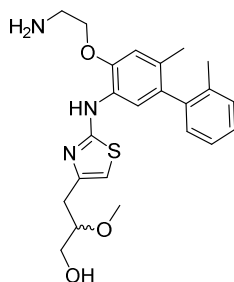
Clint was determined from the rate of test compound disappearing.

Microsomal Rat Clint Data for (±)-15 and (±)-16:

Compound	Clint (mL/hr/mg protein)
(±)-15	1.4
(±)-16	10.2

Microsomal rat Clint of (±)-**16** was very high leading to expected very high clearance in rat.

11f. Investigation of the Intravenous Pharmacokinetic Parameters of (±)-15 in the Han Wistar Rat



(±)-15

Method

The study was carried out using n=1 Han Wistar rat. The compound was administered intravenously to a fed animal at a target dose of 1 mg/kg. The actual dose was 0.83mg/kg in a dose volume: 2 mL/kg. The compound was formulated as a solution of DMSO/hydroxypropyl-beta-cyclodextrin 20% in phosphate buffer 60 mM pH 7 (5:95). Blood samples were collected at selected time points. The blood samples were then assayed using a method based upon protein precipitation with acetonitrile followed by LC/MS/MS analysis (MRM:428.4>353.3.LLQ=2ng/mL). Non-compartmental methods were used for pharmacokinetic analysis of blood concentration versus time data.

Summary of Findings

Parameters	Intravenous
Dose (mg/kg)	0.8
AUC(0-t) (ng.h/mL)	184
AUC(0-inf) (ng.h/mL)	204
T _{1/2} (h)	2.3
MRT (h)	1.7
Cl _b (mL/min/kg)	68
V _{ss} (L/kg)	6.9

12. General Chemistry and Abbreviations

General Chemistry

All solvents employed were commercially available and anhydrous grade. Reagents were used as received unless otherwise noted. All reactions were performed under a positive pressure of nitrogen or argon at ambient temperature (unless otherwise stated). Microwave-assisted reactions were run in a CEM ExplorerTM microwave. The reactions were monitored by LCMS or thin-layer chromatography (TLC) and terminated as judged by the consumption of starting material. Flash chromatography was performed using an automated Biotage[®] SP4 system with prepacked disposable SiO₂ cartridges, or using a Teledyne[®] CombiFlash ISCO HPLC with prepacked SiO₂ cartridges. Preparative HPLC was performed using an Agilent 1100 LC-MS preparative system (operating in electrospray positive ionization mode) and the columns were Waters Atlantis Prep T3 OBD 5 μ m 100 x 19 mm or a Waters SunFire Prep C18 OBD 5 μ m 100 x 19 mm, or using a Gilson reverse-phase system (liquid handler-215, UV/Vis-155, Pump-333/334) with a SunFireTM Prep C18 OBDTM 5 μ m (30 x 100 mm) column.

Proton (¹H) and carbon (¹³C{¹H}) NMR spectra were recorded in CDCl₃, CD₃OD or DMSO-*d*₆ on a Bruker Avance 400 spectrometer operating at 400 and 101 MHz, respectively. ¹H and ¹³C chemical shifts are quoted in parts per million (δ) relative to tetramethylsilane using the chemical shifts of the residual solvent peaks. ¹H NMR data are reported as follows: chemical shift (reported in parts per million), multiplicity (br = broad, d = doublet, dd = doublet of doublets, ddd = doublet of doublet of doublets, dt = doublet of triplets, m = multiplet, q = quartet, s = singlet, t = triplet, td = triplet of doublets), ¹H-¹H coupling constants (*J*) reported in hertz (Hz), integration. ¹³C chemical shifts were obtained either directly from ¹³C{¹H}

spectra or indirectly from 2D HSQC and HMBC spectra, and are referenced to δ 77.02 for CDCl_3 , δ 47.60 for CD_3OD or δ 40.05 for $\text{DMSO}-d_6$.

Analytical LCMS was conducted on either an Agilent or Shimadzu LC system. Agilent 1200 series with Mass Spec Detector coupled with an Agilent 6140 single quadrupole mass detector (operating in electrospray positive-negative switching ionization mode) and an Agilent 1200 MWD SL UV detector. LC retention times and molecular ion (m/z) are based on one of the following methods.

Agilent LCMS running conditions:

Capillary voltage: 3000V on ESI+ (2700V on ESI-)

Fragmentor/Gain: 190 on ESI+ (160 on ESI-)

Gain: 1

Drying gas flow: 12.0 L/min

Gas Temperature: 345 °C

Nebuliser Pressure: 60 psig

Scan Range: 125-800 amu

Method 1:

Eluent A: 95:5 10 mM Ammonium bicarbonate in water (pH 9.2):MeCN

Eluent B: MeCN

Gradient: 5-95% eluent B over 1.1 minutes

Flow: 0.9mL/min

Column: Waters Acquity UPLC BEH C18; 1.7 μ ; 2.1x50 mm

Column temperature: 50 °C

Method 2:

Eluent A: 97:3 10 mM Ammonium bicarbonate in water (pH 9.4):MeCN

Eluent B: MeCN

Gradient: 3.6-99% eluent B over 0.66 minutes

Flow: 0.7 mL/min

Column: YMC-Triart C18; 1.9 μ ; 2.0x50 mm

Column temperature: 45°C

Method 3: Analytical LCMS was otherwise conducted with a Shimadzu LC system with variable wavelength UV detection using reverse phase chromatography with an MeCN/water gradient and 0.02 or 0.1% TFA modifier (added to each solvent) and using a reverse phase column (Thermo Hypersil Gold C18). MS was determined using either a PE Sciex Single Quadrupole 150EC LC-MS or Waters Micromass ZQ Single Quadrupole LC-MS instrument.

The purity of final compounds was determined by analytical LCMS (as measured by UV peak area ratio) and ^1H NMR, and were determined to be >95% unless otherwise noted. HRMS was performed on an Agilent 6550 Quadrupole Time-of-Flight mass spectrometer.

Abbreviations

Aq. aqueous

(DHQD)₂PHAL 1-((*S*)-((1*R*,2*R*,4*S*,5*R*)-5-ethylquinuclidin-2-yl)(6-methoxyquinolin-4-yl)methoxy)-4-((*S*)-((1*S*,2*R*,4*S*,5*R*)-5-ethylquinuclidin-2-yl)(6-methoxyquinolin-4-yl)methoxy)phthalazine

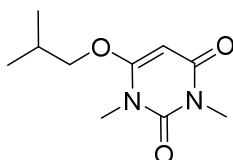
DIAD diisopropyl azodicarboxylate

DIPEA	diisopropylethylamine
DMAP	4-(dimethylamino)pyridine
ESI	electrospray ionization
HATU	1-[bis(dimethylamino)methylene]-1 <i>H</i> -1,2,3-triazolo[4,5- <i>b</i>]pyridinium 3-oxid hexafluorophosphate
HBTU	<i>N,N,N',N'</i> -tetramethyl- <i>O</i> -(1 <i>H</i> -benzotriazol-1-yl)uronium hexafluorophosphate
HPLC	high performance liquid chromatography
HRMS	high resolution mass spectrometry
IPA	propan-2-ol
MeCN	acetonitrile
Pd(dppf)Cl ₂ ·CH ₂ Cl ₂	[1,1'-bis(diphenylphosphino)ferrocene]dichloropalladium(II)·DCM
petrol	petroleum ether fraction boiling at 40-60 °C
Rochelle's salt	potassium sodium tartrate
rt	room temperature
RT	retention time
sat.	saturated
TBDMS-Cl	<i>tert</i> -butyldimethylsilyl chloride
TBDPS-Cl	<i>tert</i> -butyl(chloro)diphenylsilane
TMSCHN ₂	(trimethylsilyl)diazomethane

13. Characterization Data for Compounds 2-4

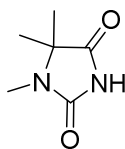
Compounds 2-4 are commercially available.

1,3-Dimethyl-6-(2-methylpropoxy)-1,2,3,4-tetrahydropyrimidine-2,4-dione (2)



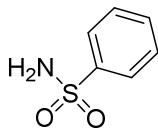
^1H NMR (CD_3OD): δ 5.21 (s, 1H), 3.92 (d, J = 6.4 Hz, 2H), 3.37 (s, 3H), 3.29 (s, 3H), 2.24-2.09 (m, 1H), 1.08 (d, J = 6.8 Hz, 6H). ^{13}C NMR (CD_3OD): δ 164.5, 162.2, 151.4, 77.2, 76.4, 27.8, 27.6, 26.8, 17.7. LCMS (method 2): m/z 213.0 $[\text{M}+\text{H}]^+$, RT = 1.26 min. Purity >95%. HRMS: Found 213.1236, $\text{C}_{10}\text{H}_{16}\text{N}_2\text{O}_3$ $[\text{M}+\text{H}]^+$ requires 213.1234 (Δ = 0.98 ppm). X-ray crystal structure gained in Lp-PLA₂.

5,5-Dimethylimidazolidine-2,4-dione (3)



^1H NMR ($\text{DMSO}-d_6$): δ 10.7 (br s, 1H), 2.73 (s, 3H), 1.27 (s, 6H). ^{13}C NMR ($\text{DMSO}-d_6$): δ 177.9, 154.9, 61.5, 23.8, 21.5. Purity >95%. HRMS: Found 143.0811, $\text{C}_6\text{H}_{10}\text{N}_2\text{O}_2$ $[\text{M} + \text{H}]^+$ requires 143.0815 (Δ = -2.26 ppm). X-ray crystal structure gained in Lp-PLA₂.

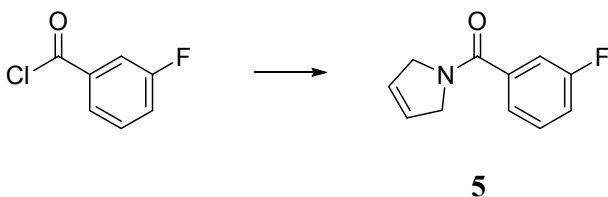
Benzenesulfonamide (4)



¹H NMR (DMSO-*d*₆): δ 7.86-7.82 (m, 2H), 7.63-7.55 (m, 3H), 7.33 (s, 2H). ¹³C NMR (DMSO-*d*₆): δ 144.1, 131.7, 128.9, 125.5. LCMS (method 2): *m/z* 175.0 [M+NH₄]⁺, RT = 1.00 min. Purity >95%. HRMS: Found 156.0124, C₆H₇NO₂S [M-H]⁻ requires 156.0125 (Δ = -0.49 ppm). X-ray crystal structure gained in Lp-PLA₂.

13. Experimental Details and Characterization Data for Compounds 5-15

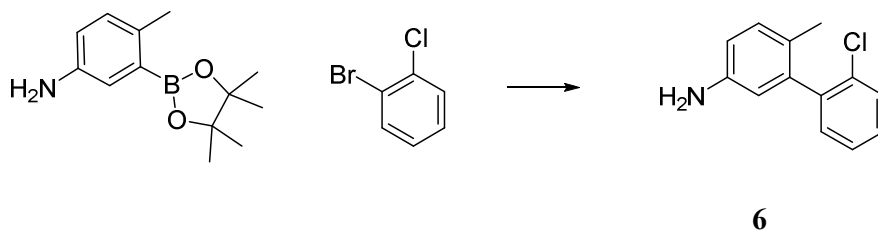
Synthesis of 1-(3-fluorobenzoyl)-2,5-dihydro-1H-pyrrole (5)



To a stirred solution of 3-fluorobenzoyl chloride (250 mg, 1.55 mmol) in DCM (7.7 mL), cooled in an ice bath, was added DIPEA (0.539 mL, 3.09 mmol) and 3-pyrroline (0.148 mL, 1.85 mmol). The mixture was allowed to warm to ambient temperature slowly and stirred for a total of 2 hours. The reaction was quenched with water and the product extracted with EtOAc (x3). The combined organic layers were washed with brine, dried over MgSO₄,

filtered, and evaporated *in vacuo*. The product was purified by preparative HPLC to yield 1-(3-fluorobenzoyl)-2,5-dihydro-1H-pyrrole (**5**) as a colourless oil (290 mg, 98%). ¹H NMR (CD₃OD): δ 7.52 (td, *J* = 8.0, 5.6 Hz, 1H), 7.39 (dt, *J* = 7.6, 1.3 Hz, 1H), 7.33 (ddd, *J* = 9.2 Hz, 2.7 Hz, 1.5 Hz, 1H), 7.29-7.22 (m, 1H), 5.99-5.95 (m, 1H), 5.88-5.83 (m, 1H), 4.43-4.39 (m, 2H), 4.28-4.24 (m, 2H). ¹³C NMR (CD₃OD): δ 169.1 (d, *J* = 2.4 Hz), 161.9 (d, *J* = 204.9 Hz), 138.5 (d, *J* = 7.2 Hz), 130.4 (d, *J* = 8.0 Hz), 125.2, 124.9, 122.4 (d, *J* = 3.2 Hz), 116.7 (d, *J* = 21.1 Hz), 113.5 (d, *J* = 23.1 Hz), 55.4, 53.1. LCMS (method 2): *m/z* 192.0 [M+H]⁺, RT = 1.25 min. Purity >95%. HRMS: Found 192.0822, C₁₁H₁₀FNO [M+H]⁺ requires 192.0819 (Δ = 1.72 ppm). X-ray crystal structure gained in Lp-PLA₂.

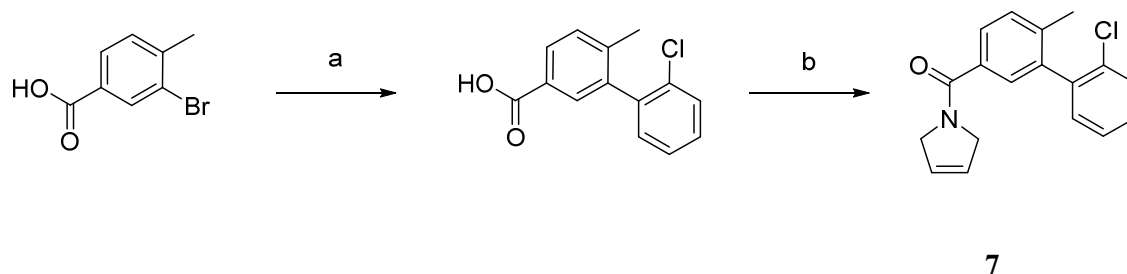
Synthesis of 2'-chloro-6-methyl-[1,1'-biphenyl]-3-amine (**6**)



To a stirred solution of 5-amino-2-methylphenylboronic acid, pinacol ester (300 mg, 1.29 mmol) in toluene:water:EtOH (4:2:1, 4 mL) was added 2-bromochlorobenzene (305 mg, 1.54 mmol) and Na₂CO₃ (409 mg, 3.86 mmol). The mixture was degassed for 5 minutes with nitrogen before Pd(PPh₃)₄ (30 mg, 0.026 mmol) was added. Degassing was continued for a further 10 minutes before heating to 120 °C (microwave) for 2 hours. The mixture was allowed to cool to ambient temperature and left for 16 hours. The reaction was diluted with water and the product was extracted with EtOAc (x2). The combined organic layers were washed with brine, dried over MgSO₄, filtered and evaporated *in vacuo*. The product was

purified by flash chromatography (gradient elution with 0-10% EtOAc:DCM) to yield 2'-chloro-6-methyl-[1,1'-biphenyl]-3-amine (**6**) as a pale yellow oil (221 mg, 79%). ¹H NMR (CD₃OD): δ 7.51-7.42 (m, 1H), 7.39-7.28 (m, 2H), 7.26-7.17 (m, 1H), 7.02 (d, *J* = 8.1 Hz, 1H), 6.71 (dd, *J* = 8.1, 2.5 Hz, 1H), 6.53 (d, *J* = 2.4 Hz, 1H), 1.95 (s, 3H). ¹³C NMR (CD₃OD): δ 144.6, 141.1, 139.9, 133.0, 130.8, 130.0, 128.9, 128.2, 126.4, 125.4, 116.5, 115.2, 17.5 LCMS (method 1): *m/z* 218.0 (³⁵Cl) and 220.0 (³⁷Cl) [M+H]⁺, RT = 1.39 min. Purity >95%. HRMS: Found 218.0732 and 220.0709, C₁₃H₁₂ClN [M+H]⁺ requires 218.0731 (³⁵Cl) (Δ = 0.09 ppm) and 220.0702 (³⁷Cl). X-ray crystal structure gained in Lp-PLA₂.

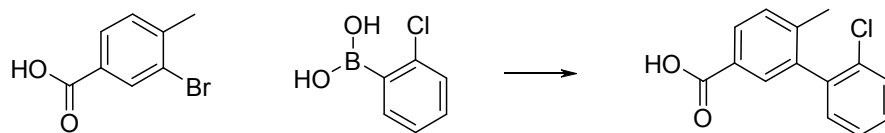
Synthesis of 1-{2'-chloro-6-methyl-[1,1'-biphenyl]-3-carbonyl}-2,5-dihydro-1H-pyrrole (7**)**



Reagents and conditions: (a) 2-Chlorophenylboronic acid, Pd(PPh₃)₄, K₂CO₃, DMF, H₂O, 110 °C, 13 h, 24% (crop 1); (b) 3-Pyrroline, HBTU, DIPEA, DCM, rt, 16 h, 58%.

Step a:

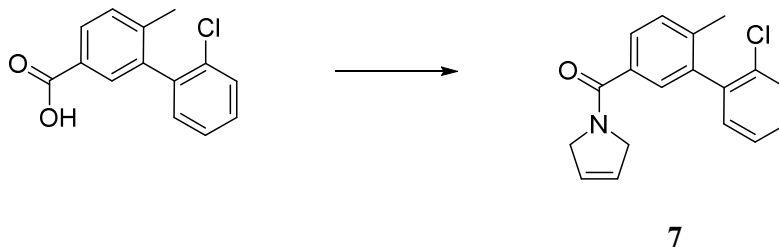
Synthesis of 2'-chloro-6-methyl-[1,1'-biphenyl]-3-carboxylic acid



3-Bromo-4-methylbenzoic acid (2.18 g, 8.62 mmol) and 2-chlorophenylboronic acid (2.02 g, 12.9 mmol) were dissolved in DMF:water (5:1, 57 mL), and K_2CO_3 (1.67 g, 12.2 mmol) was added. The stirred mixture was degassed for 10 minutes, after which $Pd(PPh_3)_4$ (199 mg, 0.172 mmol) was introduced, and the degassing was continued for a further 5 minutes. The reaction was heated at 110 °C for 6 hours and then allowed to cool. More $Pd(PPh_3)_4$ (100 mg, 0.086 mmol) was added and the heating was continued at 110 °C for a further 7 hours. The reaction was allowed to cool to ambient temperature and then diluted with water. The pH was adjusted to pH 4 with acetic acid and the product was extracted with diethyl ether (x2). The combined organic layers were washed with brine and dried over $MgSO_4$. The product was filtered and evaporated *in vacuo*. The residue was triturated with MeOH to yield a crop of 2'-chloro-6-methyl-[1,1'-biphenyl]-3-carboxylic acid as an off white solid (500 mg, 24%). The filtrate contained more product and was kept. 1H NMR ($DMSO-d_6$): δ 12.86 (s, 1H), 7.89 (dd, $J = 7.9, 1.9$ Hz, 1H), 7.65 (d, $J = 1.8$ Hz, 1H), 7.62-7.57 (m, 1H), 7.49-7.43 (m, 3H), 7.37-7.32 (m, 1H), 2.12 (s, 3H). ^{13}C NMR ($DMSO-d_6$): δ 166.9, 141.0, 138.9, 138.9, 132.0, 131.0, 130.2, 130.0, 129.6, 129.3, 128.8, 128.4, 127.4, 19.5. LCMS (method 2): m/z 245.0 (^{35}Cl) and 246.9 (^{37}Cl) $[M-H]^-$, RT = 1.11 min. Purity >95%. HRMS: Found 245.0372 and 247.0342, $C_{14}H_{11}ClO_2$ $[M-H]^-$ requires 245.0375 (^{35}Cl) ($\Delta = -1.11$ ppm) and 247.0345 (^{37}Cl).

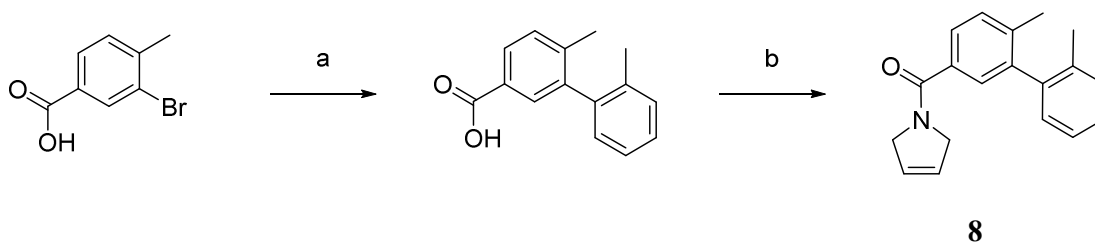
Step b:

Synthesis of 1-{2'-chloro-6-methyl-[1,1'-biphenyl]-3-carbonyl}-2,5-dihydro-1H-pyrrole (**7**)



To a stirred solution of 2'-chloro-6-methyl-[1,1'-biphenyl]-3-carboxylic acid (50 mg, 0.20 mmol) in DCM (1 mL) was added the DIPEA (71 μ L, 0.24 mmol), HBTU (94 mg, 0.24 mmol) and then 3-pyrroline (19 μ L, 0.24 mmol). The mixture was stirred at ambient temperature for 16 hours. The reaction was quenched by diluting with water and extracting the product with EtOAc (x3). The combined organic layers were washed with brine, dried over MgSO_4 , filtered, and evaporated *in vacuo*. The product was purified by preparative HPLC to yield 1-{2'-chloro-6-methyl-[1,1'-biphenyl]-3-carbonyl}-2,5-dihydro-1H-pyrrole (**7**) as a colourless oil (35 mg, 58%). ^1H NMR (CD_3OD): δ 7.56-7.50 (m, 2H), 7.44-7.38 (m, 3H), 7.32 (d, $J = 1.8$ Hz, 1H), 7.31-7.27 (m, 1H), 5.98-5.93 (m, 1H), 5.88-5.83 (m, 1H), 4.43-4.39 (m, 2H), 4.34-4.30 (m, 2H), 2.16 (s, 3H). ^{13}C NMR (CD_3OD): δ 170.4, 139.6, 139.6, 138.7, 133.7, 132.9, 130.8, 129.8, 129.2, 129.0, 127.6, 126.8, 126.2, 125.3, 125.0, 55.6, 53.1, 18.5. LCMS (method 2): m/z 298.0 (^{35}Cl) and 300.0 (^{37}Cl) $[\text{M}+\text{H}]^+$, RT = 1.45 min. Purity >95%. HRMS: Found 298.0991 and 300.0979, $\text{C}_{18}\text{H}_{16}\text{ClNO}$ $[\text{M}+\text{H}]^+$ requires 298.0993 (^{35}Cl) ($\Delta = -0.68$ ppm) and 300.0964 (^{37}Cl).

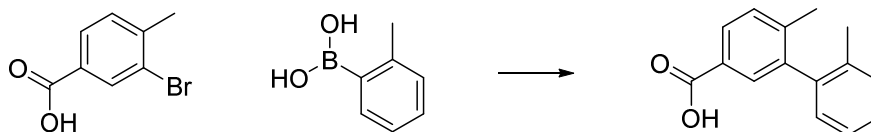
Synthesis of 1-{2',6-dimethyl-[1,1'-biphenyl]-3-carbonyl}-2,5-dihydro-1H-pyrrole (8)



Reagents and conditions: (a) 2-Methylphenylboronic acid, Pd(dppf)Cl₂·CH₂Cl₂, Na₂CO₃, 1,4-dioxane, H₂O, 90 °C, 2 h, 63%; (b) 3-Pyrroline, HATU, DIPEA, DMF, rt, 5 h, 72%.

Step a:

Synthesis of 2',6-dimethyl-[1,1'-biphenyl]-3-carboxylic acid

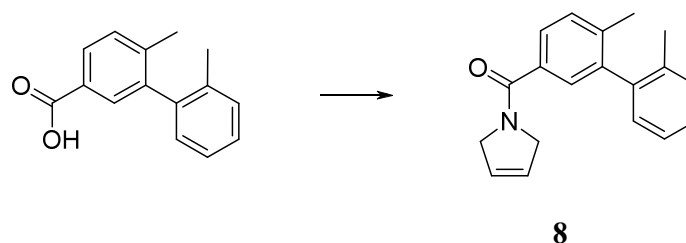


To a stirred solution of 3-bromo-4-methylbenzoic acid (360 mg, 1.63 mmol) in 1,4-dioxane:water (2:1, 8.1 mL) was added 2-methylphenylboronic acid (250 mg, 2.44 mmol) and Na₂CO₃ (345 mg, 3.26 mmol). The reaction was degassed with nitrogen for 10 minutes and then Pd(dppf)Cl₂·CH₂Cl₂ (27 mg, 0.033 mmol) was added. The mixture was heated in a Reacti-VialTM to 90 °C for 2 hours. The reaction was allowed to cool to ambient temperature and was then diluted with water. The pH was adjusted to pH 4 with acetic acid. The resulting precipitate was filtered, washed with water, and dried in a vacuum oven to yield 2',6-dimethyl-[1,1'-biphenyl]-3-carboxylic acid as a beige solid (233 mg, 63%). ¹H NMR (DMSO-*d*₆): δ 12.77 (br s, 1H), 7.86 (dd, *J* = 7.9, 1.9 Hz, 1H), 7.60 (d, *J* = 1.9 Hz, 1H), 7.45

(d, $J = 7.9$ Hz, 1H), 7.35-7.24 (m, 3H), 7.11-7.07 (m, 1H), 2.07 (s, 3H), 1.99 (s, 3H). ^{13}C NMR (DMSO- d_6): δ 167.1, 141.1, 140.7, 140.0, 134.9, 130.1, 129.9, 129.8, 128.8, 128.3, 128.2, 127.6, 125.8, 19.6, 19.3. LCMS (method 2): m/z 225.2 $[\text{M}-\text{H}]^-$, RT = 1.13 min. Purity >95%. HRMS: Found 225.0923, $\text{C}_{15}\text{H}_{14}\text{O}_2$ $[\text{M}-\text{H}]^-$ requires 225.0921 ($\Delta = 0.79$ ppm).

Step b:

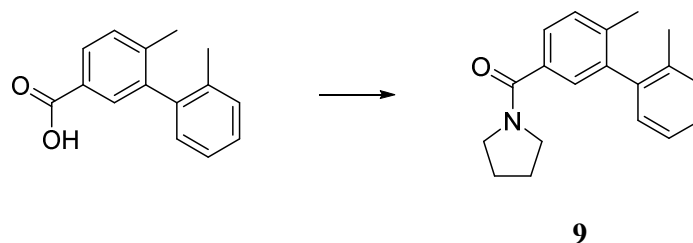
Synthesis of 1-{2',6-dimethyl-[1,1'-biphenyl]-3-carbonyl}-2,5-dihydro-1H-pyrrole (**8**)



To a stirred solution of 2',6-dimethyl-[1,1'-biphenyl]-3-carboxylic acid (100 mg, 0.44 mmol) and HATU (208 mg, 0.53 mmol) in DMF (1.5 mL) at ambient temperature, was added DIPEA (92 μL , 0.53 mmol) and then 3-pyrroline (53 μL , 0.66 mmol). The solution was stirred at ambient temperature for 5 hours. The reaction was quenched by diluting with water and extracting the product with EtOAc (x3). The combined organic layers were washed with brine, dried over MgSO_4 , filtered and evaporated *in vacuo*. The product was purified by preparative HPLC to yield 1-{2',6-dimethyl-[1,1'-biphenyl]-3-carbonyl}-2,5-dihydro-1H-pyrrole (**8**) as a pale yellow oil (88 mg, 72%). ^1H NMR (CD_3OD): δ 7.48 (dd, $J = 7.9, 1.9$ Hz, 1H), 7.41 (d, $J = 8.0$ Hz, 1H), 7.33-7.22 (m, 4H), 7.11-7.07 (m, 1H), 5.98-5.93 (m, 1H), 5.88-5.82 (m, 1H), 4.43-4.38 (m, 2H), 4.33-4.29 (m, 2H), 2.10 (s, 3H), 2.07 (s, 3H). ^{13}C NMR (CD_3OD): δ 170.6, 141.9, 140.5, 138.4, 135.3, 133.7, 129.8, 129.7, 128.7, 127.4, 127.4, 125.5, 125.5, 125.3, 125.0, 55.6, 53.1, 18.5, 18.5. LCMS (method 2): m/z 278.0 $[\text{M}+\text{H}]^+$, RT

= 1.44 min. Purity >95%. HRMS: Found 278.1538, C₁₉H₁₉NO [M+H]⁺ requires 278.1539 (Δ = -0.53 ppm). X-ray crystal structure gained in Lp-PLA₂.

Synthesis of 1-{2',6-dimethyl-[1,1'-biphenyl]-3-carbonyl}pyrrolidine (**9**)

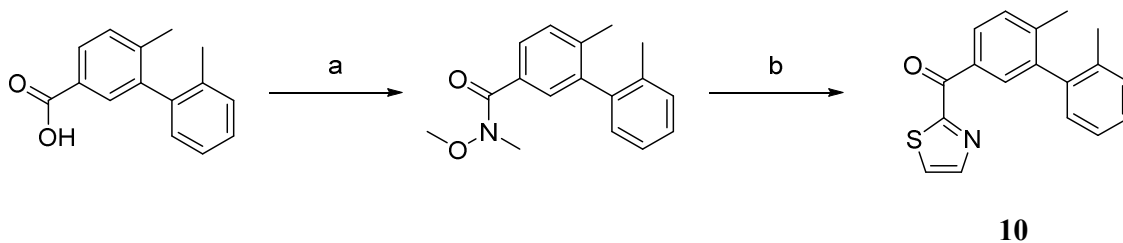


Preparation of starting material 2',6-dimethyl-[1,1'-biphenyl]-3-carboxylic acid is described within the synthesis of 1-{2',6-dimethyl-[1,1'-biphenyl]-3-carbonyl}-2,5-dihydro-1H-pyrrole (**8**) as step a.

To a solution of 2',6-dimethyl-[1,1'-biphenyl]-3-carboxylic acid (66 mg, 0.23 mmol) in DMF (1.5 mL) was added HATU (110 mg, 0.28 mmol), DIPEA (0.049 mL, 0.28 mmol) and pyrrolidine (0.030 mL, 0.35 mmol) at ambient temperature. The mixture was stirred for 2 hours and was then quenched with water. The product was extracted with EtOAc (x3). The combined organic layers were washed with brine, dried over MgSO₄, filtered, and evaporated *in vacuo*. The product was purified by preparative HPLC to yield 1-{2',6-dimethyl-[1,1'-biphenyl]-3-carbonyl}pyrrolidine (**9**) as a colourless solid (41 mg, 50%). ¹H NMR (CD₃OD): δ 7.45 (dd, J = 7.9, 1.9 Hz, 1H), 7.40 (d, J = 7.9 Hz, 1H), 7.34-7.28 (m, 2H), 7.28-7.22 (m, 2H), 7.13-7.04 (m, 1H), 3.60 (t, J = 6.8 Hz, 2H), 3.52 (t, J = 6.8 Hz, 2H), 2.09 (s, 3H), 2.06 (s, 3H), 2.05-1.96 (m, 2H), 1.96-1.86 (m, 2H). ¹³C NMR (CD₃OD): δ 170.3, 141.7, 140.5, 138.2, 135.4, 132.9, 129.7, 129.7, 128.7, 127.6, 127.3, 125.7, 125.5, 49.6, 46.1, 25.8, 23.9,

18.5, 18.5. LCMS (method 1): m/z 280.1 $[M+H]^+$, RT = 1.43 min. Purity >95%. HRMS: Found 280.1695, $C_{19}H_{21}NO$ $[M+H]^+$ requires 280.1696 (Δ = -0.12 ppm).

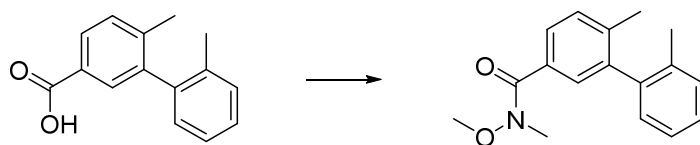
Synthesis of 2-{2',6-dimethyl-[1,1'-biphenyl]-3-carbonyl}-1,3-thiazole (10)



Reagents and conditions: (a) *N,O*-Dimethylhydroxylamine·HCl, HATU, DIPEA, DCM, rt, 16 h, 61%; (b) Thiazole, *n*-BuLi, THF, -78 °C to rt, 17.5 h, 30%.

Step a:

Synthesis of *N*-methoxy-*N*,2',6-trimethyl-[1,1'-biphenyl]-3-carboxamide



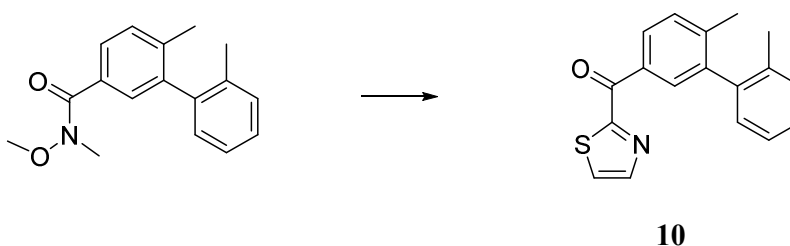
Preparation of starting material 2',6-dimethyl-[1,1'-biphenyl]-3-carboxylic acid is described within the synthesis of 1-{2',6-dimethyl-[1,1'-biphenyl]-3-carbonyl}-2,5-dihydro-1H-pyrrole (**8**) as step a.

To 2',6-dimethyl-[1,1'-biphenyl]-3-carboxylic acid (330 mg, 1.46 mmol) in DCM (7.3 mL) was added HATU (686 mg, 1.75 mmol) and then DIPEA (0.762 mL, 4.37 mmol) at ambient

temperature. *N,O*-Dimethylhydroxylamine hydrochloride (180 mg, 1.75 mmol) was added and the reaction stirred at ambient temperature for 16 hours. The reaction was quenched by addition of sat. NaHCO₃ (aq.) and the aqueous layer was extracted with EtOAc (x3). The combined organic layers were washed with brine, dried over MgSO₄, filtered and evaporated *in vacuo*. The product was purified by flash chromatography (gradient elution with 10-50% EtOAc/petrol) to give *N*-methoxy-*N*,2',6-trimethyl-[1,1'-biphenyl]-3-carboxamide as a colourless oil (241 mg, 61%). ¹H NMR (CD₃OD): δ 7.57 (dd, *J* = 7.9, 2.1 Hz, 1H), 7.40 (d, *J* = 7.9 Hz, 1H), 7.37 (d, *J* = 2.1 Hz, 1H), 7.33-7.23 (m, 3H), 7.11-7.07 (m, 1H), 3.62 (s, 3H), 3.37 (s, 3H), 2.10 (s, 3H), 2.06 (s, 3H). ¹³C NMR (CD₃OD): δ 170.2, 141.5, 140.5, 139.0, 135.3, 131.2, 129.7, 129.4, 128.7, 128.6, 127.4, 126.7, 125.5, 60.2, 32.9, 18.6, 18.5. LCMS (method 2): *m/z* 270.0 [M+H]⁺, RT = 1.43 min. Purity >95%. HRMS: Found 270.1487, C₁₇H₁₉NO₂ [M+H]⁺ requires 270.1489 (Δ = -0.51 ppm).

Step b:

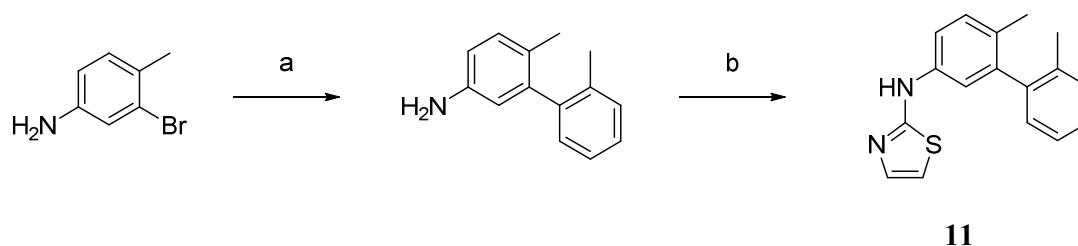
Synthesis of 2-{2',6-dimethyl-[1,1'-biphenyl]-3-carbonyl}-1,3-thiazole (**10**)



To a stirred solution of *N*-methoxy-*N*,2',6-trimethyl-[1,1'-biphenyl]-3-carboxamide (137 mg, 0.605 mmol) and thiazole (56 mg, 0.787 mmol) in THF (1.8 mL) at -78 °C was added *n*-BuLi (0.230 mL, 2.5 M, 0.575 mmol) dropwise. The mixture was allowed to warm to ambient temperature slowly and stirred for a total of 17.5 hours. The mixture was diluted with water

and the product was extracted with DCM (x3). The combined organic layers were dried over MgSO_4 , filtered and evaporated *in vacuo*. The product was purified by preparative HPLC to yield 2-{2',6-dimethyl-[1,1'-biphenyl]-3-carbonyl}-1,3-thiazole (**10**) as a colourless solid (52.7 mg, 30%). ^1H NMR (CD_3OD): δ 8.39 (dd, $J = 8.0, 1.9$ Hz, 1H), 8.24 (d, $J = 1.9$ Hz, 1H), 8.12 (d, $J = 3.1$ Hz, 1H), 8.00 (d, $J = 3.1$ Hz, 1H), 7.50 (d, $J = 8.0$ Hz, 1H), 7.36-7.25 (m, 3H), 7.15-7.11 (m, 1H), 2.17 (s, 3H), 2.09 (s, 3H). ^{13}C NMR (CD_3OD): δ 183.5, 167.7, 144.6, 142.7, 141.8, 140.4, 135.4, 132.8, 131.7, 129.8, 129.7, 129.7, 128.8, 127.4, 126.4, 125.5, 18.9, 18.6. LCMS (method 1): m/z 294.0 $[\text{M}+\text{H}]^+$, RT = 1.61 min. Purity >95%. HRMS: Found 294.0942, $\text{C}_{18}\text{H}_{15}\text{NOS}$ $[\text{M}+\text{H}]^+$ requires 294.0947 ($\Delta = -1.50$ ppm). X-ray crystal structure gained in Lp-PLA_2 .

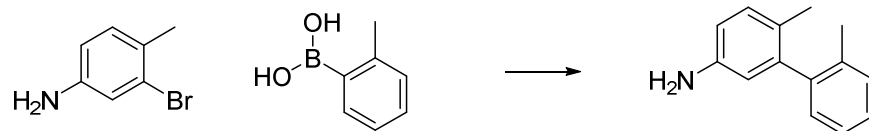
Synthesis of *N*-{2',6-dimethyl-[1,1'-biphenyl]-3-yl}-1,3-thiazol-2-amine (**11**)



Reagents and conditions: (a) 2-Methylphenylboronic acid, $\text{Pd}(\text{PPh}_3)_4$, K_2CO_3 , 1,4-dioxane, H_2O , 95 °C, 16 h, 70%; (b) 2-Bromothiazole, 37% HCl , EtOH , 90 °C, 24 h, 40%.

Step a:

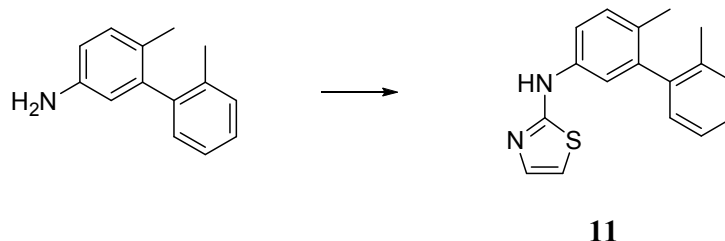
Synthesis of 2',6-dimethyl-[1,1'-biphenyl]-3-amine



To a solution of 3-bromo-4-methylaniline (300 mg, 1.58 mmol) in 1,4-dioxane:water (2:1, 7.9 mL) was added 2-methylphenylboronic acid (645 mg, 4.74 mmol) and K_2CO_3 (655 mg, 4.74 mmol). The reaction was degassed with nitrogen for 10 minutes and then $Pd(PPh_3)_4$ (37 mg, 0.032 mmol) was added. The mixture was heated at 95 °C (thermally) for 16 hours and then allowed to cool to ambient temperature. The reaction was quenched by diluting with EtOAc and the product was extracted with HCl (1 M, x2). The pH of the combined aqueous layers was adjusted with 5M NaOH to pH ~10 and the product extracted with EtOAc (x3). These combined organic layers were washed with brine, dried over $MgSO_4$, filtered and evaporated *in vacuo* to yield 2',6-dimethyl-[1,1'-biphenyl]-3-amine as a colourless solid (233 mg, 70%). 1H NMR ($DMSO-d_6$): δ 7.28-7.17 (m, 3H), 7.03-7.00 (m, 1H), 6.92 (d, $J = 8.1$ Hz, 1H), 6.49 (dd, $J = 8.1, 2.5$ Hz, 1H), 6.29 (d, $J = 2.4$ Hz, 1H), 4.89 (s, 2H), 2.01 (s, 3H), 1.81 (s, 3H). ^{13}C NMR ($DMSO-d_6$): δ 146.0, 141.9, 141.3, 134.9, 130.1, 129.5, 128.7, 126.8, 125.5, 121.7, 114.8, 113.2, 19.3, 18.4. LCMS (method 1): m/z 198.0 $[M+H]^+$, RT = 1.43 min. Purity >95%. HRMS: Found 198.1279, $C_{14}H_{15}N$ $[M+H]^+$ requires 198.1277 ($\Delta = 0.44$ ppm).

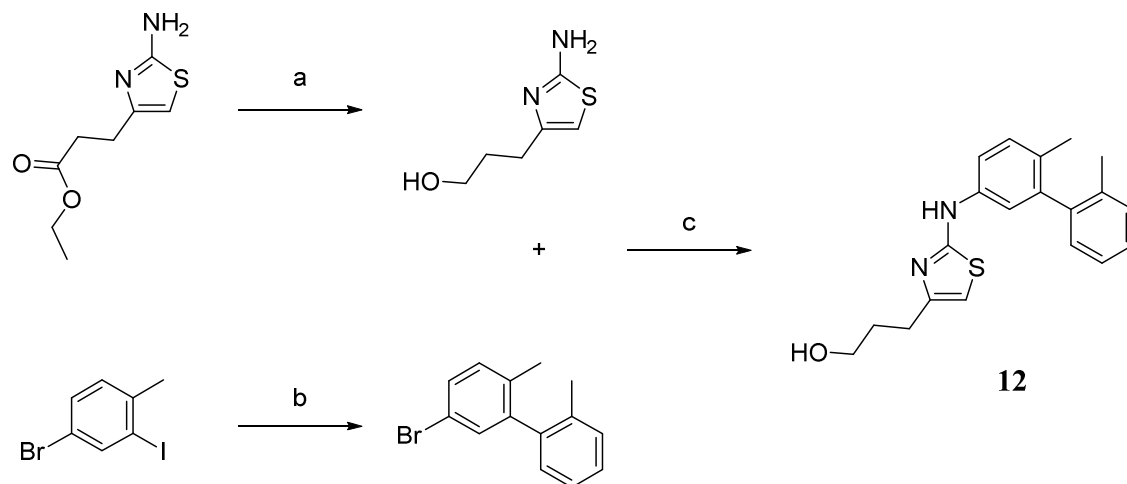
Step b:

Synthesis of *N*-{2',6-dimethyl-[1,1'-biphenyl]-3-yl}-1,3-thiazol-2-amine (**11**)



To a solution of 2',6-dimethyl-[1,1'-biphenyl]-3-amine (250 mg, 1.27 mmol) and 2-bromothiazole (0.228 mL, 2.53 mmol) in 10% aq. EtOH (5.0 mL) was added 37% HCl (0.5 mL) dropwise and the mixture was heated at 90 °C (thermally) for 24 hours. The reaction was allowed to cool to ambient temperature before being evaporated *in vacuo* and re-dissolved in EtOAc. The organic layer was washed with sat. NaHCO₃ (aq.) and dried over MgSO₄. After filtration, the solvent was evaporated *in vacuo* and the product was purified by preparative HPLC to yield *N*-{2',6-dimethyl-[1,1'-biphenyl]-3-yl}-1,3-thiazol-2-amine (**11**) as a pale brown solid (142 mg, 40%). ¹H NMR (CD₃OD): δ 7.41 (dd, *J* = 8.2, 2.5 Hz, 1H), 7.31-7.20 (m, 5H), 7.17 (d, *J* = 3.8 Hz, 1H), 7.11-7.08 (m, 1H), 6.73 (d, *J* = 3.7 Hz, 1H), 2.09 (s, 3H), 2.00 (s, 3H). ¹³C NMR (CD₃OD): δ 166.3, 142.3, 141.4, 138.9, 138.0, 135.4, 130.1, 129.5, 129.3, 128.8, 127.0, 125.3, 118.6, 116.8, 107.2, 18.6, 17.8. LCMS (method 1): *m/z* 281.0 [M+H]⁺, RT = 1.51 min. Purity >95%. HRMS: Found 281.1104, C₁₇H₁₆N₂S [M+H]⁺ requires 281.1107 (Δ = -1.00 ppm). X-ray crystal structure gained in Lp-PLA₂.

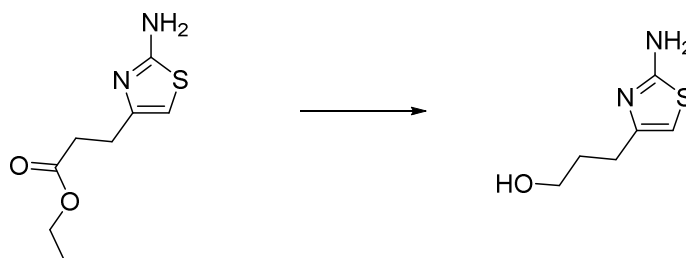
Synthesis of 3-[2-({2',6-dimethyl-[1,1'-biphenyl]-3-yl}amino)-1,3-thiazol-4-yl]propan-1-ol (12)



Reagents and conditions: (a) LiAlH_4 , THF, 0-5 °C, 1 h, 48%; (b) 2-Methylphenylboronic acid, $\text{Pd}(\text{PPh}_3)_4$, K_2CO_3 , toluene, H_2O , 85 °C, 16 h, 98%; (c) CuI , K_2CO_3 , toluene, 100 °C, 20 h, 29%.

Step a:

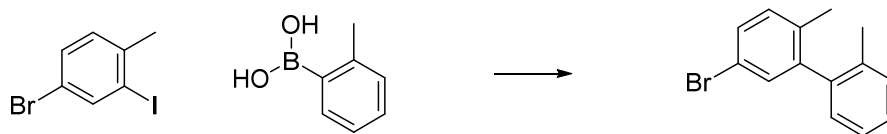
Synthesis of 3-(2-amino-1,3-thiazol-4-yl)propan-1-ol



Ethyl 3-(2-amino-1,3-thiazol-4-yl)propanoate (300 mg, 1.61 mmol) in THF (8.1 mL) was cooled in an ice-water bath. Powdered LiAlH_4 (122 mg, 3.22 mmol) was added in small portions. The reaction was stirred for 1 hour and quenched carefully with water (1 mL). Rochelles salt (v:v ~1:2 in water) was added and the product extracted with DCM:IPA (3:1, x4). The combined organic layers were washed with brine and dried over MgSO_4 . After filtration, the product was purified by preparative HPLC to yield 3-(2-amino-1,3-thiazol-4-yl)propan-1-ol as a colourless solid (122 mg, 48%). ^1H NMR ($\text{DMSO}-d_6$): δ 6.74 (s, 2H), 6.08 (s, 1H), 4.40 (s, 1H), 3.41 (br t, $J = 6.5$ Hz, 2H), 2.41 (t, $J = 7.6$ Hz, 2H), 1.73-1.65 (m, 2H). ^{13}C NMR (CD_3OD): δ 167.0, 151.1, 100.6, 60.9, 31.3, 27.2. LCMS (method 2): m/z 159.0 $[\text{M}+\text{H}]^+$, RT = 0.99 min. Purity >95%. HRMS: Found 159.0587, $\text{C}_6\text{H}_{10}\text{N}_2\text{OS}$ $[\text{M}+\text{H}]^+$ requires 159.0587 ($\Delta = -0.03$ ppm).

Step b:

Synthesis of 5-bromo-2,2'-dimethyl-1,1'-biphenyl

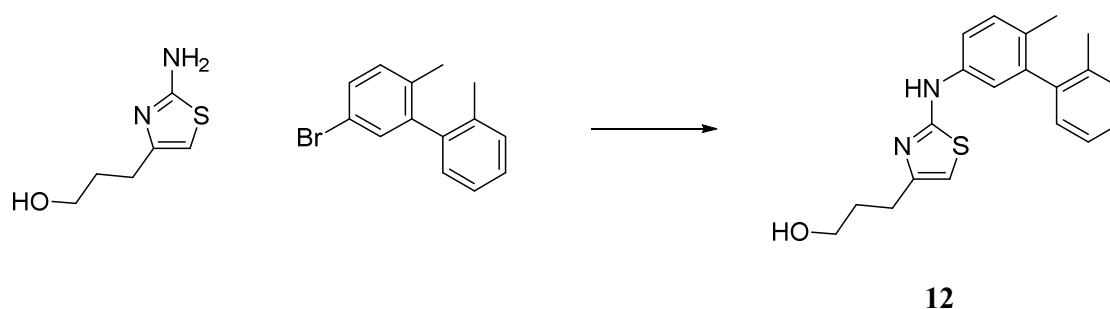


To a solution of 4-bromo-2-iodo-1-methylbenzene (250 mg, 0.84 mmol) in toluene:water (1:1, 4.2 mL) was added 2-methylphenylboronic acid (115 mg, 0.84 mmol) and K_2CO_3 (175 mg, 1.27 mmol). The reaction was degassed with nitrogen for 10 minutes and then $\text{Pd}(\text{PPh}_3)_4$ (49 mg, 0.04 mmol) was added. The mixture was heated at 85 °C (thermally) for 16 hours. The reaction was allowed to cool to ambient temperature and then diluted with water. The product was extracted with EtOAc (x3). The combined aqueous layers were washed with

brine, dried over MgSO_4 , filtered and evaporated *in vacuo*. The product was purified by flash chromatography (100% petrol) to yield 5-bromo-2,2'-dimethyl-1,1'-biphenyl as a colourless oil (216 mg, 98%). ^1H NMR (CD_3OD): δ 7.41 (dd, $J = 8.2, 2.2$ Hz, 1H), 7.32-7.19 (m, 5H), 7.07-7.03 (m, 1H), 2.05 (s, 3H), 1.99 (s, 3H). ^{13}C NMR (CD_3OD): δ 143.8, 140.1, 135.2, 134.9, 131.5, 131.3, 129.9, 129.7, 128.6, 127.4, 125.5, 118.6, 18.4, 18.0. LCMS (method 2): m/z none $[\text{M}+\text{H}]^+$, RT = 1.74 min. Purity >95%.

Step c:

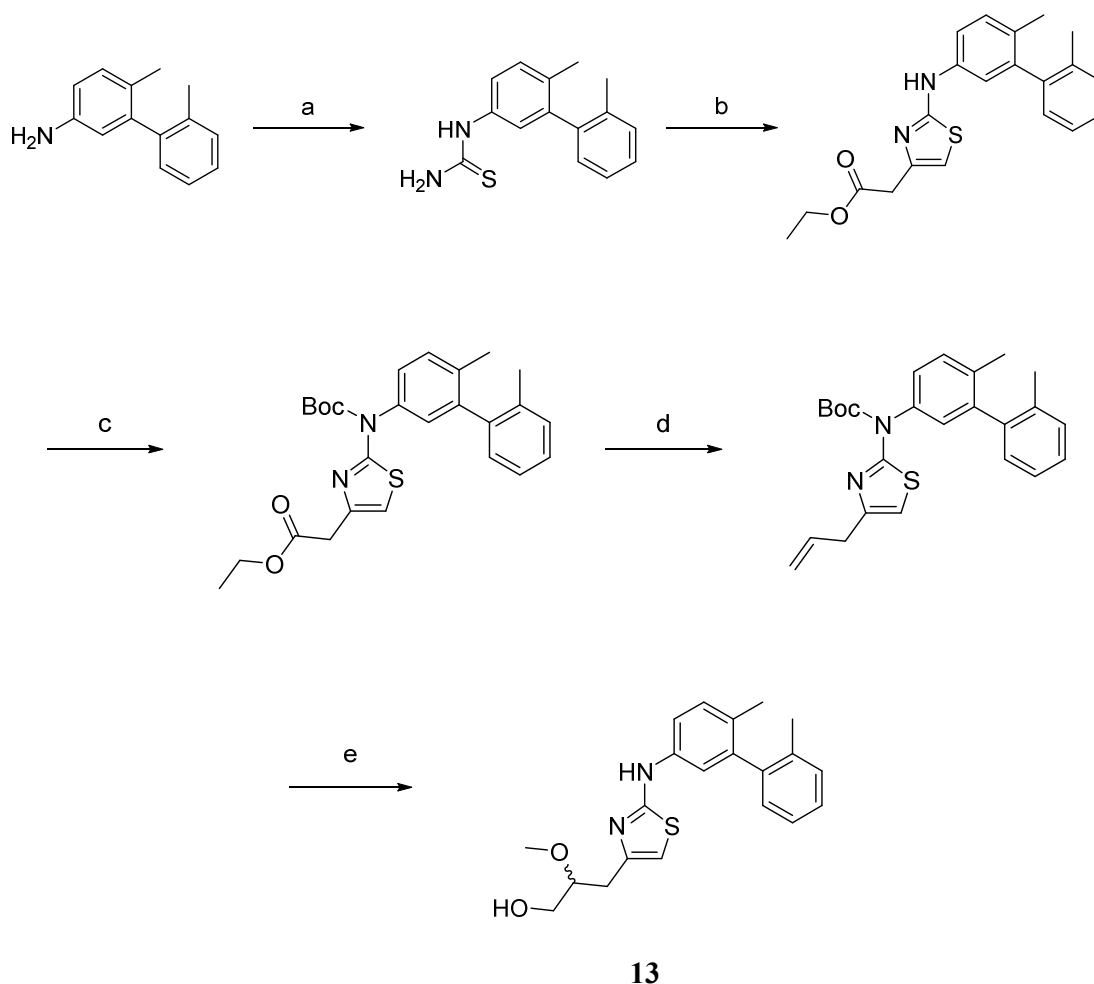
Synthesis of 3-[2-(2',6-dimethyl-[1,1'-biphenyl]-3-yl)amino]-1,3-thiazol-4-yl]propan-1-ol (12)



In a Reacti-VialTM was added 3-(2-amino-1,3-thiazol-4-yl)propan-1-ol (58 mg, 0.402 mmol), CuI (2.6 mg, 0.013 mmol) and K_2CO_3 (77.8 mg, 0.563 mmol). The vessel was back filled with nitrogen several times before 5-bromo-2,2'-dimethyl-1,1'-biphenyl (70 mg, 0.286 mmol) in toluene (2.0 mL) was introduced under a stream of nitrogen. The sealed vial was heated to 100 °C (thermally) for 20 hours. The reaction was allowed to cool to ambient temperature and was diluted with petrol. The mixture was filtered through a bed of Celite® and washed further with EtOAc (x3). The combined organic layers were evaporated *in vacuo*. The product was purified by preparative HPLC to yield 3-[2-(2',6-dimethyl-[1,1'-biphenyl]-3-yl)amino]-1,3-thiazol-4-yl]propan-1-ol.

yl}amino)-1,3-thiazol-4-yl]propan-1-ol (**12**) as a colourless oil (25 mg, 29%). ¹H NMR (CD₃OD): δ 7.45 (dd, *J* = 8.3, 2.5 Hz, 1H), 7.31-7.20 (m, 4H), 7.16 (d, *J* = 2.5 Hz, 1H), 7.11-7.07 (m, 1H), 6.31 (s, 1H), 3.61 (t, *J* = 6.5 Hz, 2H), 2.68-2.62 (m, 2H), 2.10 (s, 3H), 1.99 (s, 3H), 1.94-1.86 (m, 2H). LCMS (method 2): *m/z* 339.0 [M+H]⁺, RT = 1.44 min. Purity >95%. HRMS: Found 339.1517, C₂₀H₂₂N₂OS [M+H]⁺ requires 339.1526 (Δ = -2.78 ppm). X-ray crystal structure gained in Lp-PLA₂.

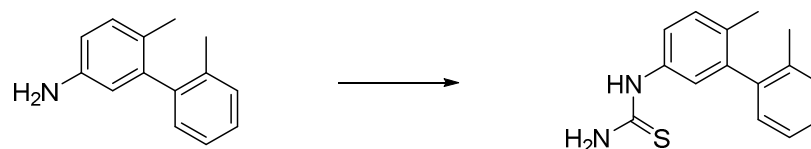
Synthesis of 3-[2-({2',6-dimethyl-[1,1'-biphenyl]-3-yl}amino)-1,3-thiazol-4-yl]-2-methoxypropan-1-ol (13**)**



Reagents and conditions: (a) 1-(1H-Imidazole-1-carbothioyl)-1H-imidazole, DCM, rt, 3 h, then NH₄OH, rt, 18 h, 99%; (b) Methyl 4-chloro-3-oxobutanoate, EtOH, 80 °C, 16 h, 98%; (c) Boc₂O, DMAP, DCM, 23 °C, 2 h, 58%; (d) DIBAL-H, toluene, DCM, -78 °C, 2 h; then methyltriphenylphosphonium bromide, KO^tBu, THF, 0 °C, 30 min, 39% (over 2 steps); (e) (DHQD)₂PHAL, *tert*-BuOH, H₂O, 23 °C, 16 h; then imidazole, TBDMS-Cl, DCM, 0 °C to 23 °C, 1 h, 52%; then Ag₂O, MeI, MeCN, 60 °C, 16 h, 72%; then 4 M HCl, 23 °C, 16 h, 86%.

Step a:

Synthesis of {2',6-dimethyl-[1,1'-biphenyl]-3-yl}thiourea



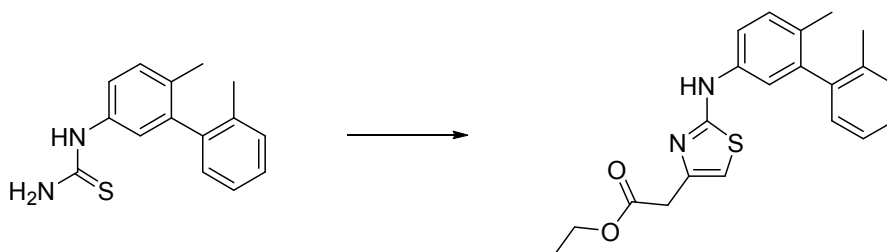
Preparation of starting material 2',6-dimethyl-[1,1'-biphenyl]-3-amine is described within the synthesis of *N*-{2',6-dimethyl-[1,1'-biphenyl]-3-yl}-1,3-thiazol-2-amine (**11**) as step a.

To a solution of 2',6-dimethyl-[1,1'-biphenyl]-3-amine (10.6 g, 53.7 mmol) in DCM (100 mL) was added 1-(1H-imidazole-1-carbothioyl)-1H-imidazole (11.49 g, 64.5 mmol). The reaction was stirred at ambient temperature for 3 hours, before ammonium hydroxide (30 mL, 53.7 mmol) was added. The mixture was stirred for a further 18 hours at ambient temperature. The solvent was evaporated *in vacuo*, and water added. The precipitate was filtered to yield {2',6-dimethyl-[1,1'-biphenyl]-3-yl}thiourea (13.7 g, 99%). ¹H NMR

(CD₃OD): δ 7.30-7.20 (m, 5H), 7.08-6.99 (m, 2H), 2.04 (s, 3H), 2.01 (s, 3H). LCMS (method 3): m/z 257.2 [M+H]⁺, RT = 1.09 min. Purity >95%.

Step b:

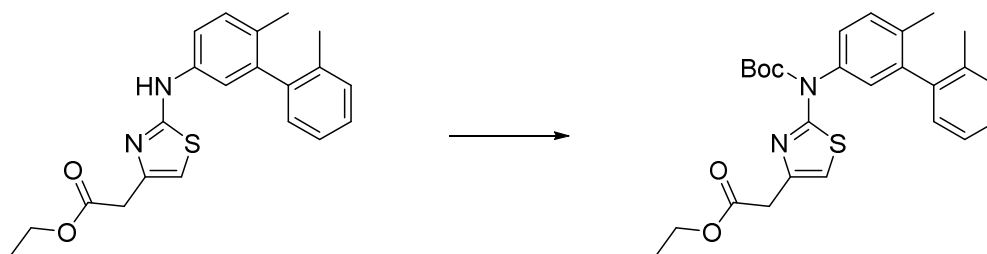
Synthesis of ethyl 2-[2-({2',6-dimethyl-[1,1'-biphenyl]-3-yl}amino)-1,3-thiazol-4-yl]acetate



To a solution of {2',6-dimethyl-[1,1'-biphenyl]-3-yl}thiourea (13.7 g, 53.4 mmol) in EtOH (100 mL) was added methyl 4-chloro-3-oxobutanoate (8.05 g, 53.4 mmol). The mixture was heated to 80 °C (thermally) for 16 hours and allowed to cool to ambient temperature. The solvent was evaporated *in vacuo*. Water (100 mL) was added to the residue and the pH was adjusted to pH 8 with sat. NaHCO₃ (aq.). The product was extracted with DCM (3x100 mL) and the combined organic layers were dried over Na₂SO₄. After filtration, the solvent was evaporated *in vacuo* to yield ethyl 2-[2-({2',6-dimethyl-[1,1'-biphenyl]-3-yl}amino)-1,3-thiazol-4-yl]acetate (18.5 g, 98%). ¹H NMR (CDCl₃): δ 7.30-7.20 (m, 5H), 7.11 (d, J = 6.8 Hz, 1H), 7.03 (d, J = 2.4 Hz, 1H), 6.43 (s, 1H), 4.19 (q, J = 7.2 Hz, 2H), 3.72 (s, 2H), 2.09 (s, 3H), 2.02 (s, 3H), 1.27 (t, J = 7.2 Hz, 3H). LCMS (method 3): m/z 367.0 [M+H]⁺, RT = 1.35 min. Purity >85%.

Step c:

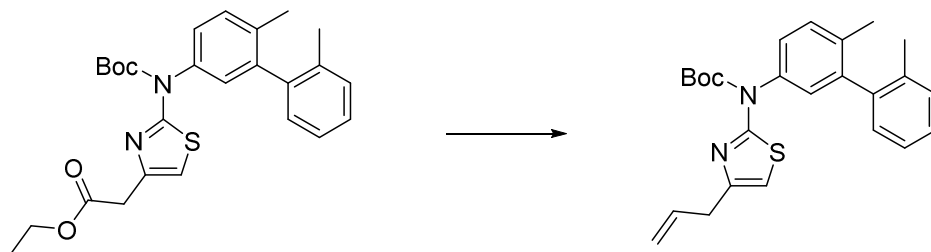
Synthesis of ethyl 2-(2-({[(*tert*-butoxy)carbonyl]({2',6-dimethyl-[1,1'-biphenyl]-3-yl})amino}-1,3-thiazol-4-yl)acetate



To a solution of ethyl 2-[2-(({2',6-dimethyl-[1,1'-biphenyl]-3-yl}amino)-1,3-thiazol-4-yl)acetate (2.0 g, 5.67 mmol) in DCM (20 mL) was added Boc_2O (1.58 mL, 6.81 mmol) and DMAP (0.069 g, 0.567 mmol). The reaction mixture was stirred at 23 °C for 2 hours. The solvent was evaporated *in vacuo* and the product was purified by flash chromatograph (petrol:EtOAc from 20:1 to 15:1) to give ethyl 2-(2-({[(*tert*-butoxy)carbonyl]({2',6-dimethyl-[1,1'-biphenyl]-3-yl})amino}-1,3-thiazol-4-yl)acetate as a brown oil (1.5 g, 58%). ^1H NMR (CDCl_3): δ 7.30 (d, $J = 8.0$ Hz, 1H), 7.26-7.20 (m, 3H), 7.15 (d, $J = 6.8$ Hz, 1H), 7.12 (dd, $J = 8.0, 2.4$ Hz, 1H), 6.98 (d, $J = 2.0$ Hz, 1H), 6.81 (s, 1H), 4.12 (q, $J = 6.8$ Hz, 2H), 3.65 (s, 2H), 2.10 (s, 3H), 2.09 (s, 3H), 1.44 (s, 9H), 1.20 (t, $J = 6.8$ Hz, 3H). LCMS (method 3): m/z 467.2 $[\text{M}+\text{H}]^+$, RT = 1.39 min. Purity >95%.

Step d:

Synthesis of *tert*-butyl *N*-{2',6-dimethyl-[1,1'-biphenyl]-3-yl}-*N*-[4-(prop-2-en-1-yl)-1,3-thiazol-2-yl]carbamate

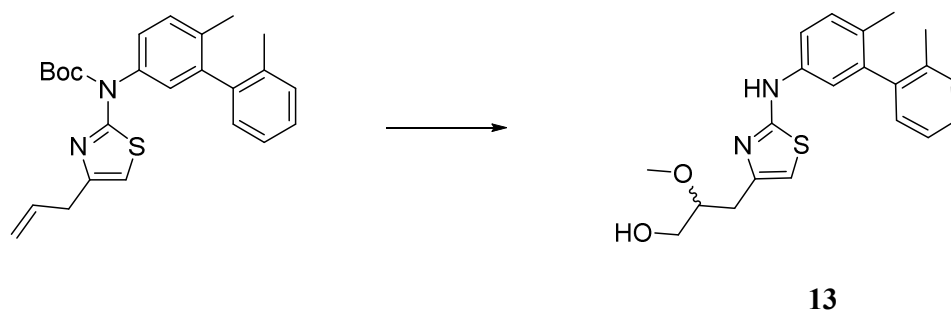


To a stirred solution of ethyl 2-(2-((*tert*-butoxy)carbonyl)({2',6-dimethyl-[1,1'-biphenyl]-3-yl})amino)-1,3-thiazol-4-yl)acetate (850 mg, 1.88 mmol) in DCM (5 mL) and toluene (5 mL) at -78 °C, was added a solution of DIBAL-H (2.25 mL, 1 M, 2.25 mmol) dropwise. The reaction mixture was stirred at -78 °C for 2 hours and was then quenched with acetone (1.0 mL) at -78 °C. Stirring was continued for 10 minutes before the addition of 10% aqueous tartaric acid (10 mL). The mixture was allowed to warm to ambient temperature and the product was extracted with EtOAc (50 mL). The organic phase was washed with brine (20 mL), dried over Na₂SO₄, filtered and evaporated *in vacuo* to yield *tert*-butyl *N*-{2',6-dimethyl-[1,1'-biphenyl]-3-yl}-*N*-[4-(2-oxoethyl)-1,3-thiazol-2-yl]carbamate as a yellow oil which was used directly for next step. LCMS (method 3): *m/z* 423.0 [M+H]⁺, RT = 1.37 min. Purity 85%. To a stirred solution of methyltriphenylphosphonium bromide (2.01 g, 5.64 mmol) in THF (30 mL) at 0 °C was added KO^{*t*}Bu (633 mg, 5.64 mmol) portionwise. The reaction mixture was stirred at 0 °C for 30 minutes. To this reaction mixture was added a solution of *tert*-butyl *N*-{2',6-dimethyl-[1,1'-biphenyl]-3-yl}-*N*-[4-(2-oxoethyl)-1,3-thiazol-2-yl]carbamate (794 mg, 1.88 mmol) in THF (5 mL) dropwise at 0 °C. The mixture was allowed to warm to ambient temperature and stirred for a total of 2 hours. The reaction

mixture was partitioned between EtOAc (50 mL) and water (20 mL). The organic phase was separated, dried over Na₂SO₄, filtered and evaporated *in vacuo*. The product was purified by flash chromatography (gradient elution with EtOAc/petrol) to yield *tert*-butyl *N*-{2',6-dimethyl-[1,1'-biphenyl]-3-yl}-*N*-[4-(prop-2-en-1-yl)-1,3-thiazol-2-yl]carbamate (300 mg, 39% two steps). ¹H NMR (CDCl₃): δ 7.31 (d, *J* = 8.0 Hz, 1H), 7.26-7.20 (m, 3H), 7.17-7.13 (m, 2H), 6.98 (d, *J* = 2.0 Hz, 1H), 6.55 (s, 1H), 5.99-5.89 (m, 1H), 5.10-5.04 (m, 2H), 3.32 (d, *J* = 6.8 Hz, 2H), 2.10 (s, 3H), 2.01 (s, 3H), 1.45 (s, 9H). LCMS (method 3): *m/z* 421.2 [M+H]⁺, RT = 1.44 min. Purity >95%.

Step e:

Synthesis of 3-[2-({2',6-dimethyl-[1,1'-biphenyl]-3-yl}amino)-1,3-thiazol-4-yl]-2-methoxypropan-1-ol (**13**)



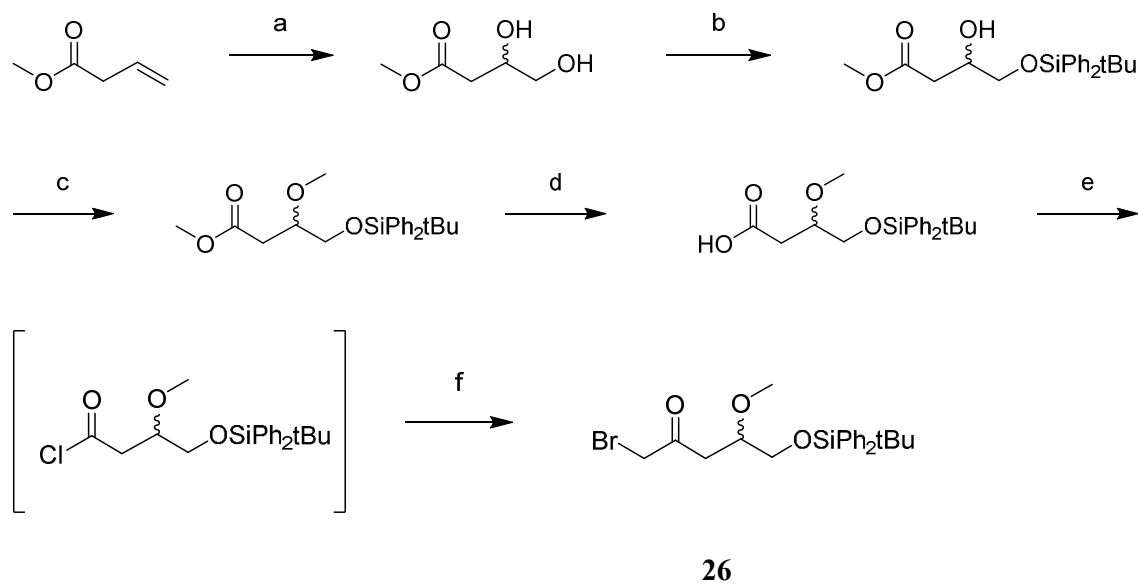
To a solution of *tert*-butyl *N*-{2',6-dimethyl-[1,1'-biphenyl]-3-yl}-*N*-[4-(prop-2-en-1-yl)-1,3-thiazol-2-yl]carbamate (100 mg, 0.238 mmol) in water (5 mL) and *tert*-BuOH (10 mL) at 23 °C was added solid (DHQD)₂PHAL (1.0 g, 1.28 mmol). The reaction mixture was stirred at 23 °C for 16 hours. The reaction mixture was partitioned between EtOAc (50 mL) and water (25 mL). The organic phase was separated, dried over Na₂SO₄, filtered and evaporated *in vacuo* to yield *tert*-butyl *N*-[4-(2,3-dihydroxypropyl)-1,3-thiazol-2-yl]-*N*-{2',6-dimethyl-[1,1'-

biphenyl]-3-yl}carbamate. LCMS (method 3): m/z 455.2 $[M+H]^+$, RT = 1.29 min. Purity >75%. To a stirred solution of *tert*-butyl *N*-[4-(2,3-dihydroxypropyl)-1,3-thiazol-2-yl]-*N*-{2',6-dimethyl-[1,1'-biphenyl]-3-yl}carbamate (50 mg, 0.110 mmol) and imidazole (30.0 mg, 0.440 mmol) in DCM (1 mL) at 0 °C was added a solution of TBDMS-Cl (33.2 mg, 0.220 mmol) in DCM (1 mL) dropwise. The mixture was stirred at 23 °C for 1 hour. The reaction mixture was partitioned between DCM (20 mL) and water (10 mL). The separated organic layer was dried over Na₂SO₃, filtered and evaporated *in vacuo*. The product was purified by preparative TLC plate (petrol:EtOAc 8:1) to yield *tert*-butyl *N*-(4-{3-[(*tert*-butyldimethylsilyl)oxy]-2-hydroxypropyl}-1,3-thiazol-2-yl)-*N*-{2',6-dimethyl-[1,1'-biphenyl]-3-yl}carbamate (40 mg, 52%). LCMS (method 3): m/z 569.1 $[M+H]^+$, RT = 1.66 min. Purity >85%. To a stirred solution of *tert*-butyl *N*-(4-{3-[(*tert*-butyldimethylsilyl)oxy]-2-hydroxypropyl}-1,3-thiazol-2-yl)-*N*-{2',6-dimethyl-[1,1'-biphenyl]-3-yl}carbamate (80 mg, 0.141 mmol) in MeCN (2 mL) was added Ag₂O (130 mg, 0.563 mmol) and MeI (399 mg, 2.81 mmol). The reaction mixture was heated at 60 °C for 16 hours and allowed to cool to ambient temperature. The mixture was filtered and the filtrate was evaporated *in vacuo*. The product was purified by preparative TLC plate (petrol:EtOAc 10:1) to yield *tert*-butyl *N*-(4-{3-[(*tert*-butyldimethylsilyl)oxy]-2-methoxypropyl}-1,3-thiazol-2-yl)-*N*-{2',6-dimethyl-[1,1'-biphenyl]-3-yl}carbamate (60 mg, 72%). LCMS (method 3): m/z 583.1 $[M+H]^+$, RT = 1.46 min. Purity >95%. To *tert*-butyl *N*-(4-{3-[(*tert*-butyldimethylsilyl)oxy]-2-methoxypropyl}-1,3-thiazol-2-yl)-*N*-{2',6-dimethyl-[1,1'-biphenyl]-3-yl}carbamate (60 mg, 0.103 mmol) was added 4 M HCl (5 mL, 20.0 mmol). The reaction mixture was stirred at 23 °C for 16 hours. The mixture was evaporated *in vacuo* and the product purified by preparative HPLC to yield 3-[2-(2',6-dimethyl-[1,1'-biphenyl]-3-yl)amino]-1,3-thiazol-4-yl]-2-methoxypropan-1-ol (**13**) (33 mg, 86%). ¹H NMR (CD₃OD): δ 7.48 (d, J = 8.0 Hz, 1H), 7.38 (dd, J = 8.4, 2.4 Hz, 1H), 7.39-7.32 (m, 3H), 7.17 (d, J = 2.8 Hz, 1H), 7.11 (d, J = 7.2 Hz, 1H), 6.72 (s, 1H), 3.70-

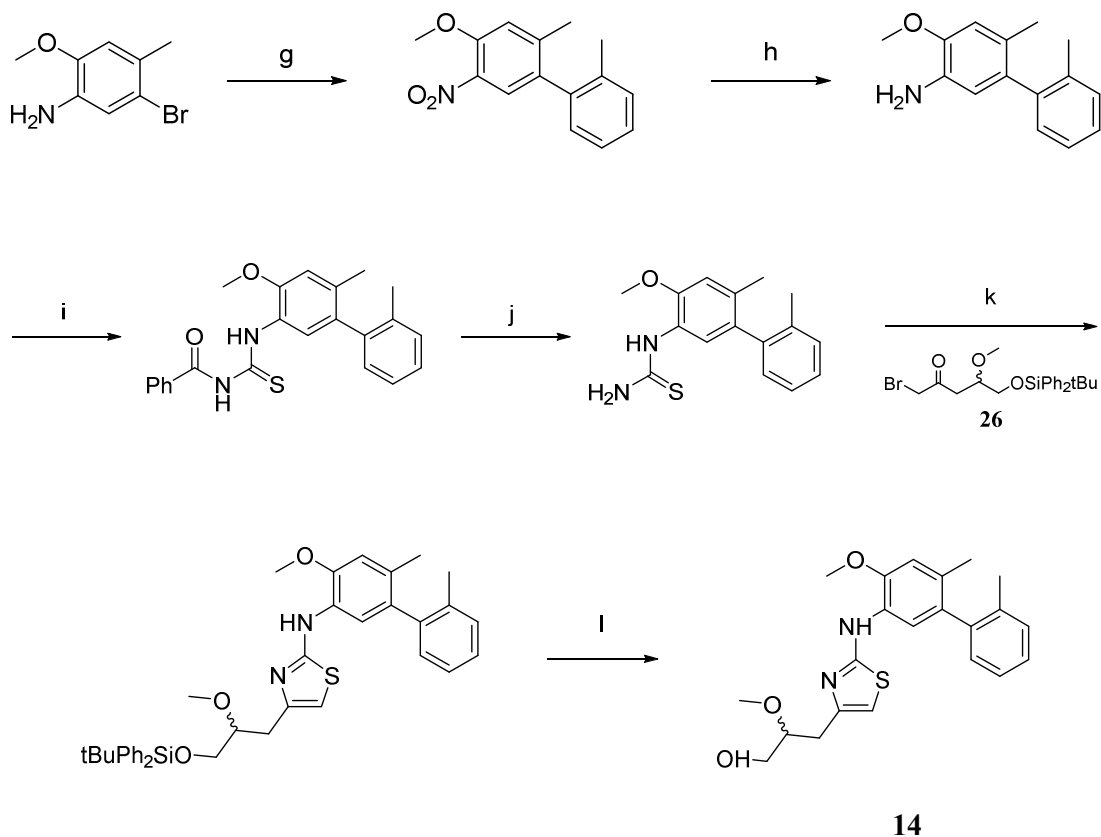
3.55 (m, 3H), 3.43 (s, 3H), 2.96 (dd, $J = 15.2, 4.4$ Hz, 1H), 2.87 (dd, $J = 15.2, 7.2$ Hz, 1H), 2.10 (s, 6H). LCMS (method 3): m/z 369.2 $[M+H]^+$, RT = 0.99 min. Purity >95%.

Synthesis of 2-methoxy-3-[2-({4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-yl}amino)-1,3-thiazol-4-yl]propan-1-ol (14)

Synthesis of intermediate 1-bromo-5-[(*tert*-butyldiphenylsilyl)oxy]-4-methoxypentan-2-one (26)



Reagents and conditions: (a) OsO_4 , NMO, acetone, H_2O , rt, 16 h, 93%; (b) TBDPS-Cl, NEt_3 , DMAP, DCM, rt, 48 h, 77%; (c) Me_3OBF_4 , proton sponge, DCM, rt, 3.5 h, 74%; (d) LiOH , THF, H_2O , rt, 24 h, 99%; (e) $(\text{COCl})_2$, DMF, DCM, 0 °C to rt, 1 h; (f) TMSCHN_2 , HBr , MeCN, 0 °C to rt, 25 min, 53% (over 2 steps).

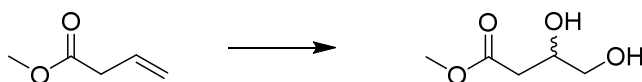


Reagents and conditions: (g) 2-Methylphenylboronic acid, Pd(dppf)Cl₂·CH₂Cl₂, Na₂CO₃, 1,4-dioxane, H₂O, 95 °C, 5 h, 88%; (h) H₂, Pd/C, EtOAc, MeOH, rt, 5 h, 91%; (i) Benzoyl isothiocyanate, DCM, rt, 3 h, 83%; (j) 1M NaOH, EtOH, 16 h, rt, 83%; (k) Bromide **26**, THF, 61 °C, 16 h, 16%; (l) TBAF, THF, rt, 3 h, 40%.

Synthesis of intermediate 1-bromo-5-[(*tert*-butyldiphenylsilyl)oxy]-4-methoxypentan-2-one
(26)

Step a:

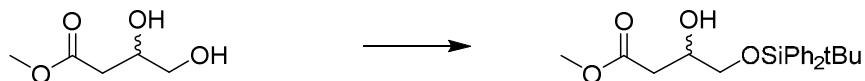
Synthesis of methyl 3,4-dihydroxybutanoate



Methyl but-3-enoate (10 g, 100 mmol) in acetone (50 mL) was added dropwise over 24 hours to a stirring mixture of NMO (12.87 g, 110 mmol) and osmium tetroxide (0.254 g, 0.99 mmol) in acetone (150 mL) and water (100 mL) at ambient temperature. The reaction was stirred for 16 hours and was then evaporated *in vacuo*. The residue was filtered through a silica-Celite® bed (250 mL of SiO₂ and 50 mL of Celite®) washing with 75% EtOAc in hexanes (1 L) and then EtOAc (1 L). The combined filtrate was evaporated *in vacuo* to yield methyl 3,4-dihydroxybutanoate (12.5 g, 93%) which was used without further purification. ¹H NMR (CDCl₃): δ 4.15 (br s, 1H), 3.75 (s, 3H), 3.75-3.67 (m, 1H), 3.60-3.52 (m, 1H), 3.34 (s, 1H), 2.62-2.51 (m, 2H), 2.35 (br s, 1H). Purity 85%.

Step b:

Synthesis of methyl 4-[(*tert*-butyldiphenylsilyl)oxy]-3-hydroxybutanoate

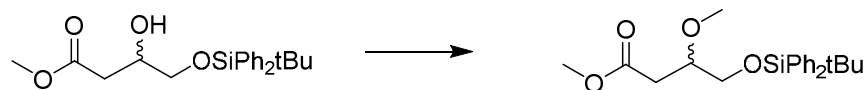


To a stirred solution of methyl 3,4-dihydroxybutanoate (11.6 g, 86.0 mmol, 85% pure) in DCM (300 mL) was added DMAP (1.06 g, 8.65 mmol) and triethylamine (14.5 mL, 104

mmol). After 10 minutes, TBDPS-Cl (23.8 mL, 86.0 mmol) was added. The mixture was stirred at ambient temperature for 48 hours. The reaction was poured into brine (500 mL) and the product extracted with DCM (x2). The combined organic layers were dried over Na₂SO₄, filtered, and evaporated *in vacuo*. The product was purified by flash chromatography (eluting with 3-12% EtOAc:hexanes) to yield methyl 4-[(*tert*-butyldiphenylsilyl)oxy]-3-hydroxybutanoate (24.7 g, 77%). ¹H NMR (CDCl₃): δ 7.69-7.67 (m, 4H), 7.47-7.40 (m, 6H), 4.25-4.15 (m, 1H), 3.72 (s, 3H), 3.72-3.63 (m, 1H), 2.90 (d, *J* = 4.5 Hz, 1H), 2.59-2.56 (m, 2H), 0.99 (s, 9H). Purity >90%.

Step c:

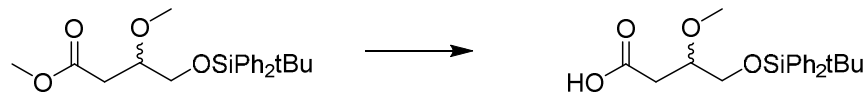
Synthesis of methyl 4-[(*tert*-butyldiphenylsilyl)oxy]-3-methoxybutanoate



To a stirred solution of methyl 4-[(*tert*-butyldiphenylsilyl)oxy]-3-hydroxybutanoate (23 g, 61.7 mmol) in DCM (750 mL) was added proton sponge (23.82 g, 111 mmol) and then Me₃OBf₄ (15.98 g, 108 mmol). The reaction was stirred for 3.5 hours at ambient temperature. The mixture was evaporated *in vacuo*, filtered and the product was purified by flash chromatography to yield methyl 4-[(*tert*-butyldiphenylsilyl)oxy]-3-methoxybutanoate as a yellow oil (17.7 g, 74%). ¹H NMR (CDCl₃): δ 7.61 (d, *J* = 4.0 Hz, 4H), 7.37-7.31 (m, 6H), 3.68-3.56 (m, 6H), 3.28 (s, 3H), 2.58-2.59 (m, 1H), 2.50-2.48 (m, 1H), 0.99 (s, 9H). Purity >95%.

Step d:

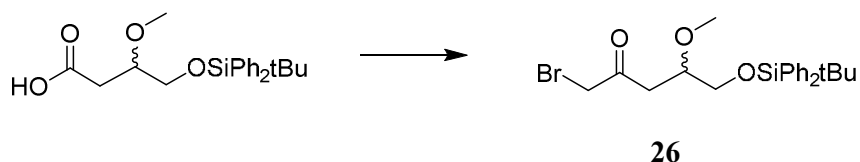
Synthesis of 4-[(*tert*-butyldiphenylsilyl)oxy]-3-methoxybutanoic acid



To a stirred solution of methyl 4-[(*tert*-butyldiphenylsilyl)oxy]-3-methoxybutanoate (6.5 g, 16.8 mmol) in THF (120 mL) and distilled water (40 mL) was added LiOH (803 mg, 33.5 mmol). The mixture was stirred at ambient temperature for 24 hours. The reaction was diluted with HCl (1 N) until pH ~3 and the product was extracted with diethyl ether (x3). The combined organic layers were dried over Na₂SO₄, filtered and evaporated *in vacuo* to yield 4-[(*tert*-butyldiphenylsilyl)oxy]-3-methoxybutanoic acid (6.20 g, 99%) which was used in the next step without purification. ¹H NMR (CDCl₃): δ 10.50 (br s, 1H), 7.70 (d, *J* = 7.3 Hz, 4H), 7.46-7.40 (m, 6H), 3.78-3.75 (m, 2H), 3.75-3.68 (m, 1H), 3.39 (s, 3H), 2.73 (dd, *J* = 15.9, 4.0 Hz, 1H), 2.62 (dd, *J* = 15.9, 7.3 Hz, 1H), 0.98 (s, 9H). Purity >95%.

Step e and f:

Synthesis of 1-bromo-5-[(*tert*-butyldiphenylsilyl)oxy]-4-methoxypentan-2-one (**26**)

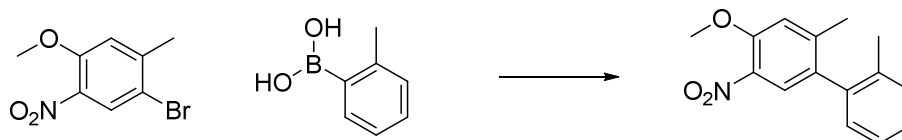


4-[(*tert*-Butyldiphenylsilyl)oxy]-3-methoxybutanoic acid (6.4 g, 17.2 mmol) was dissolved in DCM (80 mL). DMF (two drops) was added followed by oxalyl chloride (3.01 mL, 34.4 mmol) dropwise. The reaction was stirred at 0 °C for 1 hour, after which the ice-water bath

was removed and stirring continued for an additional hour. The mixture was concentrated *in vacuo* and then dissolved in MeCN (200 mL). The solution was cooled in an ice-water and TMS-diazomethane (25.8 mL, 51.5 mmol) was slowly added over 10 minutes. The reaction was allowed to warm to ambient temperature and stirred for a further 2 hours. After re-cooling in an ice-water bath, 33% HBr in AcOH (5.65 mL, 34.4 mmol) was slowly added over a 10 minute period. Stirring was continued at ambient temperature for 25 minutes. The reaction was concentrated *in vacuo* and taken up in EtOAc (250 mL). The organic layer was washed with sat. NaHCO₃ (aq.), brine and dried over Na₂SO₄. After filtration, the solvent was evaporated *in vacuo* and the product was purified by flash chromatography (eluting with 5 to 20% EtOAc/Hexane) to yield 1-bromo-5-[(*tert*-butyldiphenylsilyl)oxy]-4-methoxypentan-2-one (**26**) as a light yellow oil (4.05 g, 53%). ¹H NMR (CDCl₃): δ 7.70-7.69 (m, 4H), 7.47-7.41 (m, 6H), 3.98 (s, 2H), 3.73-3.66 (m, 3H), 3.33 (s, 3H), 2.91-2.88 (m, 2H), 1.09 (s, 9H). LCMS (method 3): *m/z* no ion [M+H]⁺, RT = 1.50 min. Purity >95%.

Step g:

Synthesis of 4-methoxy-2,2'-dimethyl-5-nitro-1,1'-biphenyl

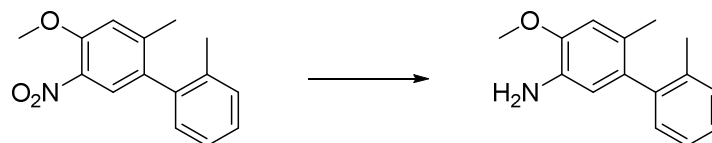


To 1-bromo-4-methoxy-2-methyl-5-nitrobenzene (500 mg, 2.03 mmol), 2-methylphenylboronic acid (331 mg, 2.43 mmol) and Na₂CO₃ (431 mg, 4.06 mmol) was added 1,4-dioxane:water (2:1, 10.2 mL). The reaction was degassed with nitrogen for 10 minutes and then Pd(dppf)Cl₂·CH₂Cl₂ (33 mg, 0.020 mmol) was added. The reaction was

heated at 95 °C for 5 hours, allowed to cool to ambient temperature, and then the solvent was evaporated *in vacuo*. To the residue was added water and the product was extracted with EtOAc (x3). The combined organic layers were washed with brine and dried over MgSO₄. After filtration, and evaporation *in vacuo*, the product was purified by flash chromatography (gradient elution with 0-20% EtOAc/petrol) to yield 4-methoxy-2,2'-dimethyl-5-nitro-1,1'-biphenyl as a colourless solid (461 mg, 88%). ¹H NMR (DMSO-*d*₆): δ 7.58 (s, 1H), 7.35 (s, 1H), 7.34-7.31 (m, 2H), 7.30-7.24 (m, 1H), 7.13-7.09 (m, 1H), 3.97 (s, 3H), 2.09 (s, 3H), 2.03 (s, 3H). LCMS (method 2): *m/z* 258.0 [M+H]⁺, RT = 1.52 min. Purity >90%.

Step h:

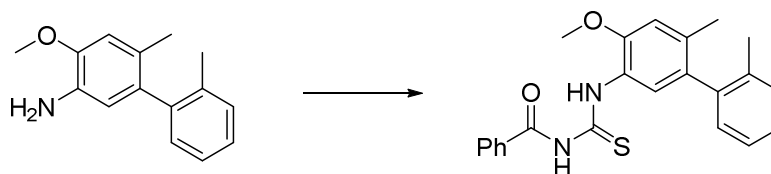
Synthesis of 4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-amine



1-Methoxy-5-methyl-4-(2-methylphenyl)-2-nitrobenzene (461 mg, 1.79 mmol) was taken up in MeOH:EtOAc (2:1, 17.9 mL) and 10% Pd/C (191 mg) was added. The reaction was shaken under an atmosphere of hydrogen, at ambient temperature for 5 hours. The reaction was filtered and the filtrate was evaporated *in vacuo* to yield 4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-amine as a colourless oil (368 mg, 91%). ¹H NMR (DMSO-*d*₆): δ 7.28-7.23 (m, 1H), 7.23-7.16 (m, 2H), 7.01 (dd, *J* = 6.9, 1.9 Hz, 1H), 6.75 (s, 1H), 6.40 (s, 1H), 5.12 (s, 2H), 3.80 (s, 3H), 2.01 (s, 3H), 1.87 (s, 3H). ¹³C NMR (CD₃OD): δ 147.5, 141.6, 135.8, 134.0, 132.0, 129.3, 129.2, 126.7, 126.6, 125.1, 117.3, 111.9, 54.8, 18.6, 18.1. LCMS (method 2): *m/z* 228.0 [M+H]⁺, RT = 1.48 min. Purity >90%. HRMS: Found 228.1385, C₁₅H₁₇NO [M+H]⁺ requires 228.1383 (Δ = 0.76 ppm).

Step i:

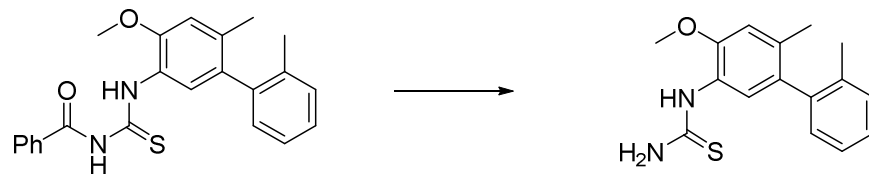
Synthesis of 1-benzoyl-3-{4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-yl}thiourea



To a stirred solution of 4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-amine (219 mg, 0.96 mmol) in DCM (5 mL) was added benzoyl isothiocyanate (0.132 mL, 0.96 mmol). The reaction was stirred at ambient temperature for 3 hours. The crude product was purified directly by flash chromatography (gradient elution with 0-30% EtOAc in petrol) to yield 1-benzoyl-3-{4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-yl}thiourea as a pale yellow solid (312 mg, 83%). ^1H NMR (CDCl_3): δ 12.76 (br s, 1H), 9.01 (br s, 1H), 8.44 (s, 1H), 7.93-7.88 (m, 2H), 7.66-7.61 (m, 1H), 7.56-7.50 (m, 2H), 7.29-7.26 (m, 1H), 7.26-7.19 (m, 2H), 7.16-7.12 (m, 1H), 6.87 (s, 1H), 3.99 (s, 3H), 2.13 (s, 3H), 2.09 (s, 3H). ^{13}C NMR (CDCl_3): δ 176.7, 166.6, 150.0, 140.9, 136.5, 134.9, 133.6, 133.5, 132.2, 130.0, 130.0, 129.3, 127.6, 127.3, 125.7, 124.7, 124.5, 112.3, 56.3, 20.3, 20.2. LCMS (method 2): m/z 391.0 $[\text{M}+\text{H}]^+$, RT = 1.63 min. Purity >90%. HRMS: Found 391.1480, $\text{C}_{23}\text{H}_{22}\text{N}_2\text{O}_2\text{S}$ $[\text{M}+\text{H}]^+$ requires 391.1475 (Δ = 1.35 ppm).

Step j:

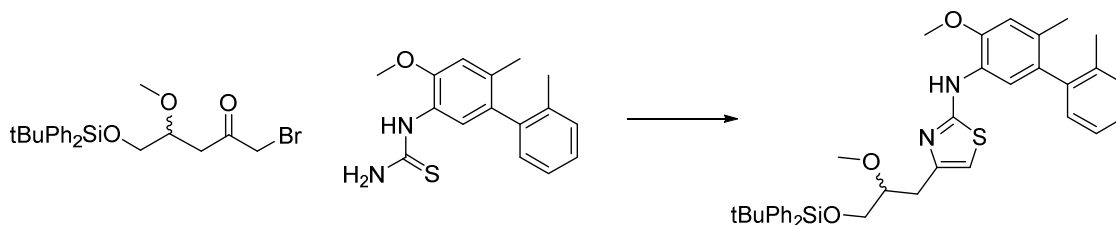
Synthesis of {4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-yl}thiourea



To a stirred solution of 1-benzoyl-3-{4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-yl}thiourea (259 mg, 0.66 mmol) in EtOH (4 mL) was added 1 M NaOH (2.7 mL, 2.66 mmol). The reaction was stirred at ambient temperature for 16 hours. The organic solvent was evaporated *in vacuo* and the residue was partitioned between EtOAc and sat. NaHCO₃ (aq.). The organic layer was separated and further extracted with EtOAc (x2). The combined organic layers were washed with brine dried over MgSO₄. After filtration, the solvent was evaporated *in vacuo* and the product purified by flash chromatography (gradient elution with 0-50% EtOAc/petrol) to yield {4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-yl}thiourea (157 mg, 83%). ¹H NMR (CDCl₃): δ 7.81 (br s, 1H), 7.29-7.20 (m, 3H), 7.08-7.05 (m, 2H), 6.88 (s, 1H), 6.24 (br s, 2H), 3.91 (s, 3H), 2.08 (s, 3H), 2.07 (s, 3H). ¹³C NMR (CDCl₃): δ 181.9, 151.6, 139.9, 136.5, 136.0, 134.6, 130.2, 129.7, 127.7, 125.8, 125.8, 123.0, 113.7, 56.0, 20.2, 20.0. LCMS (method 2): *m/z* 286.9 [M+H]⁺, RT = 1.36 min. Purity 95%. HRMS: Found 287.1219, C₁₆H₁₈N₂OS [M+H]⁺ requires 287.1213 (Δ = 2.05 ppm).

Step k:

Synthesis of 4-{3-[(*tert*-butyldiphenylsilyl)oxy]-2-methoxypropyl}-*N*-{4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-yl}-1,3-thiazol-2-amine

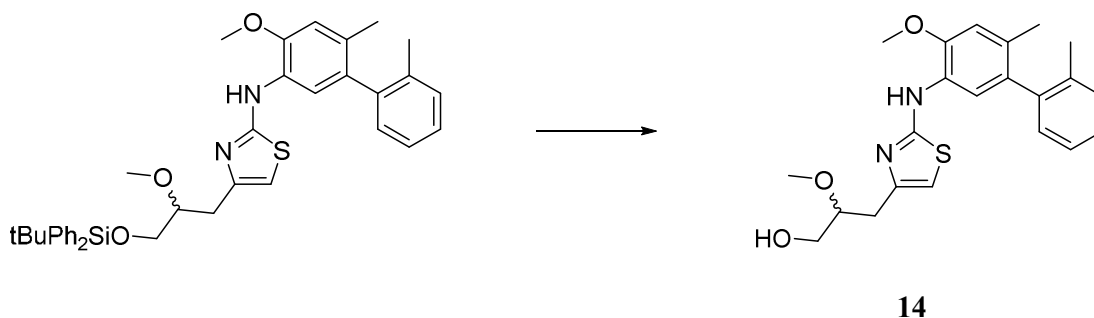


26

To a stirred solution of 1-bromo-5-[(*tert*-butyldiphenylsilyl)oxy]-4-methoxypentan-2-one **26** (162.2 mg, 0.361 mmol) in THF (5 mL) was added {4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-yl}thiourea (103 mg, 0.361 mmol). The reaction mixture was heated to 61 °C for 16 hours and then allowed to cool to ambient temperature. The solvent was then evaporated *in vacuo*. The residue was purified via flash chromatography to yield 4-{3-[(*tert*-butyldiphenylsilyl)oxy]-2-methoxypropyl}-*N*-{4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-yl}-1,3-thiazol-2-amine (37.7 mg, 16%). ¹H NMR (CD₃OD): δ 7.71 (s, 1H), 7.70-7.59 (m, 4H), 7.40-7.33 (m, 6H), 7.23-7.09 (m, 3H), 6.94 (m, 1H), 6.23 (s, 1H), 6.23 (s, 1H), 3.95 (s, 3H), 3.69-3.65 (m, 2H), 3.62-2.60 (m, 1H), 3.33 (s, 3H), 2.72 (s, 2H), 2.10 (s, 1.5H), 2.08 (s, 1.5H), 2.03 (s, 3H), 1.02 (s, 9H). LCMS (method 3): *m/z* 637.5 [M+H]⁺, RT = 0.46 min. Purity >95%.

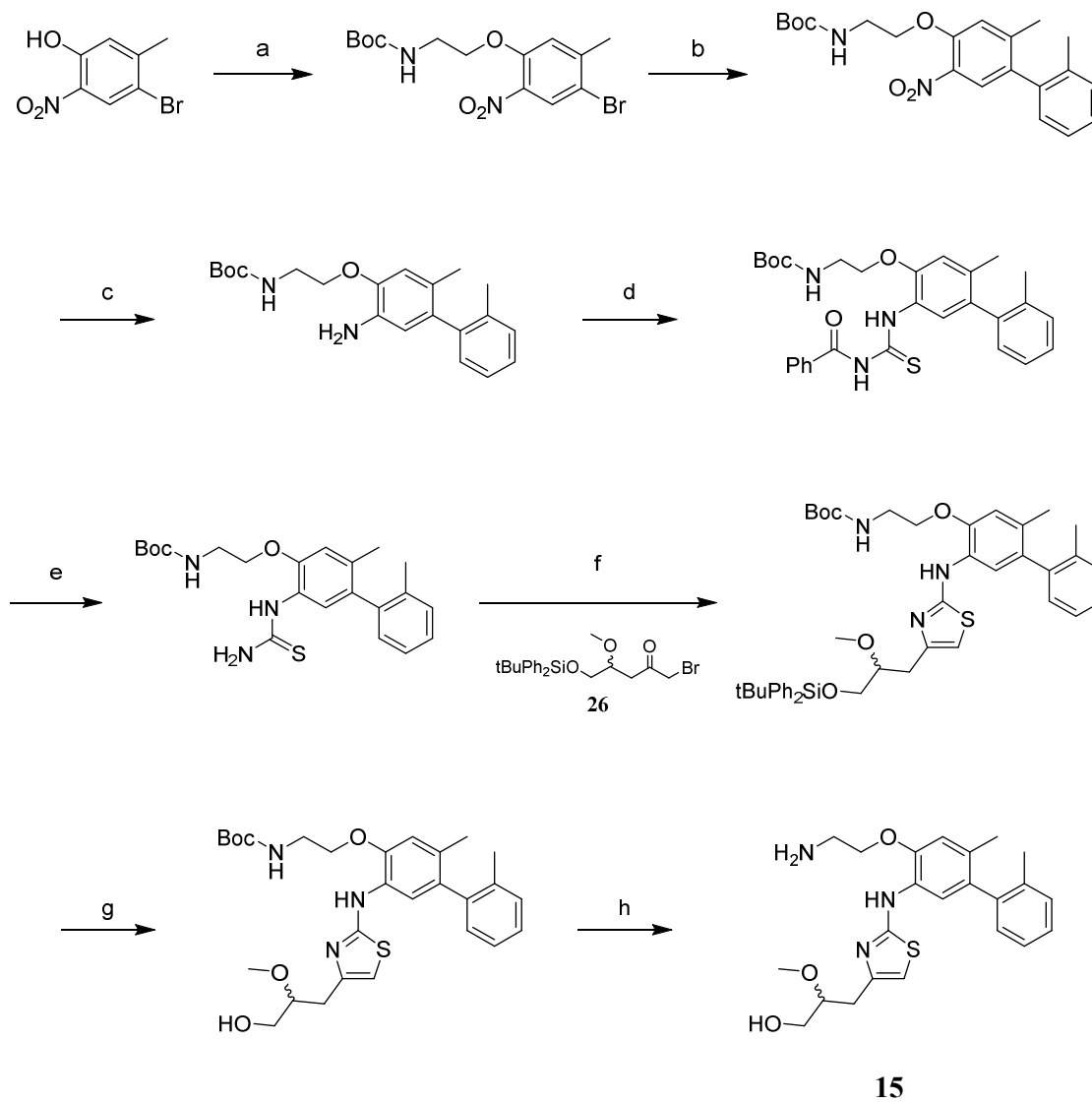
Step 1:

Synthesis of 2-methoxy-3-[2-({4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-yl}amino)-1,3-thiazol-4-yl]propan-1-ol (**14**)



To a stirred solution of 4-{3-[(*tert*-butyldiphenylsilyl)oxy]-2-methoxypropyl}-*N*-{4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-yl}-1,3-thiazol-2-amine (37.7 mg, 0.059 mmol) in THF (2 mL) was added TBAF (0.178 mL, 0.178 mmol, 1 M in THF). The mixture was stirred for 3 hours at ambient temperature and then the solvent was evaporated *in vacuo*. The residue was taken up in DCM (10 mL), washed with water (10 mL), and the organic layer was purified directly by flash chromatography to afford 2-methoxy-3-[2-({4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-yl}amino)-1,3-thiazol-4-yl]propan-1-ol (**14**) as a yellow brown solid (9.9 mg, 40%). ¹H NMR (CD₃OD): δ 7.66 (s, 1H), 7.28-7.19 (m, 3H), 7.10 (d, *J* = 8.0 Hz, 1H), 6.94 (s, 1H), 6.37 (s, 1H), 3.95 (s, 3H), 3.63-3.57 (m, 2H), 3.49-3.45 (m, 1H), 3.32 (s, 3H), 2.79-2.72 (m, 2H), 2.11 (s, 3H), 2.03 (s, 3H). LCMS (method 3): *m/z* 389.2 [M+H]⁺, RT = 0.95 min. Purity 95%.

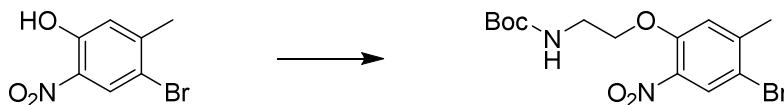
Synthesis of 3-(2-{[4-(2-aminoethoxy)-2',6-dimethyl-[1,1'-biphenyl]-3-yl]amino}-1,3-thiazol-4-yl)-2-methoxypropan-1-ol (15)



Reagents and conditions: (a) *tert*-Butyl *N*-(2-hydroxyethyl)carbamate, DIAD, PPh₃, THF, rt, 2 h, 99%; (b) 2-Methylphenylboronic acid, Pd(PPh₃)₄, Cs₂CO₃, 1,4-dioxane, H₂O, 110 °C, 1 h, 78%; (c) H₂, 10% Pd/C, EtOH, rt, 97%; (d) Benzoyl isothiocyanate, DCM, rt, 3 h, 100%; (e) 1 M NaOH, EtOH, rt, 16 h, 73%; (f) Bromide (**26**), THF, 61 °C, 3 h; (g) TBAF, THF, rt, 3 h, 75% (over 2 steps); (h) 4 M HCl in 1,4-dioxane, rt, 1 h, 95%.

Step a:

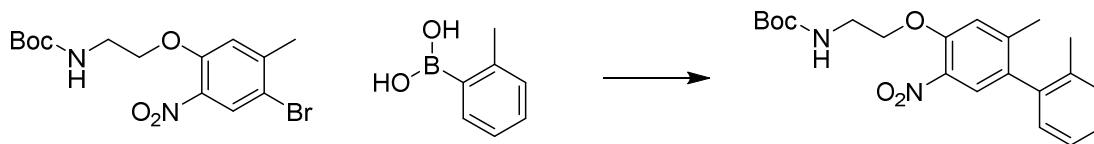
Synthesis of *tert*-butyl *N*-[2-(4-bromo-5-methyl-2-nitrophenoxy)ethyl]carbamate



To a stirred solution of 4-bromo-5-methyl-2-nitrophenol (500 mg, 2.15 mmol), DIAD (1.08 mL, 5.39 mmol) and *tert*-butyl *N*-(2-hydroxyethyl)carbamate (0.434 mL, 2.80 mmol) in THF (11 mL) was added triphenylphosphine (1.41 g, 5.39 mmol). The solution was allowed to stir for 2 hours at ambient temperature, after which the mixture was evaporated *in vacuo*. The product was purified by flash chromatography (gradient elution with 0-20% EtOAc/petrol) to yield *tert*-butyl *N*-[2-(4-bromo-5-methyl-2-nitrophenoxy)ethyl]carbamate as a colourless oil (795 mg, 99%). ¹H NMR (CD₃OD): δ 8.05 (s, 1H), 7.29 (s, 1H), 4.20 (t, *J* = 5.6 Hz, 2H), 3.47 (t, *J* = 5.6 Hz, 2H), 2.47 (s, 3H), 1.45 (s, 9H). ¹³C NMR (CD₃OD): δ 157.0, 151.1, 145.1, 138.2, 128.2, 117.1, 113.8, 79.0, 68.6, 39.3, 27.3, 22.0. LCMS (method 2): *m/z* 318.8 (⁷⁹Br) and 321.0 (⁸¹Br) [M-(*t*Bu)+H]⁺, and 373.0 (⁷⁹Br) and 375.0 (⁸¹Br) [M-H]⁻, RT = 1.50 min. Purity >95%. HRMS: Found 397.0370 and 399.0353, C₁₄H₁₉BrN₂O₅ [M+Na]⁺ requires 397.0370 (⁷⁹Br) (Δ = -0.21 ppm) and 399.0349 (⁸¹Br).

Step b:

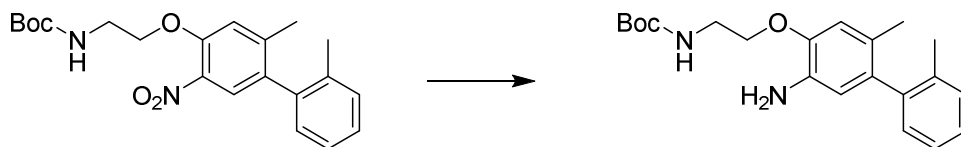
Synthesis of *tert*-butyl *N*-[2-({2,2'-dimethyl-5-nitro-[1,1'-biphenyl]-4-yl}oxy)ethyl]carbamate



To a mixture of *tert*-butyl *N*-[2-(4-bromo-5-methyl-2-nitrophenoxy)ethyl]carbamate (809 mg, 2.16 mmol), 2-methylphenylboronic acid (352 mg, 2.59 mmol) and Cs₂CO₃ (1405 mg, 4.31 mmol) was added 1,4-dioxane (13 mL) and water (2.6 mL). Argon was bubbled through the mixture and then Pd(PPh₃)₄ (49.8 mg, 0.043 mmol) was added. The reaction was heated at 110 °C (microwave) for 1 hour. The mixture was evaporated *in vacuo* and the residue was taken up in EtOAc (75 mL). The organic layer was washed with water (60 mL), brine (60 mL) and dried over MgSO₄. After filtration, the product was purified by flash chromatography (gradient elution with 0 to 30% EtOAc/hexanes) to yield *tert*-butyl *N*-[2-({2,2'-dimethyl-5-nitro-[1,1'-biphenyl]-4-yl}oxy)ethyl]carbamate (651 mg, 78%). ¹H NMR (DMSO-*d*₆): δ 7.57 (s, 1H), 7.36-7.30 (m, 3H), 7.30-7.27 (m, 1H), 7.11 (d, *J* = 7.0 Hz, 1H), 6.69 (br s, 1H), 4.21 (t, *J* = 5.5 Hz, 2H), 3.36-3.33 (m, 2H), 2.07 (s, 3H), 2.03 (s, 3H), 1.40 (s, 9H). LCMS (method 3): *m/z* 387.1 [M+H]⁺, RT = 1.33 min. Purity >95%.

Step c:

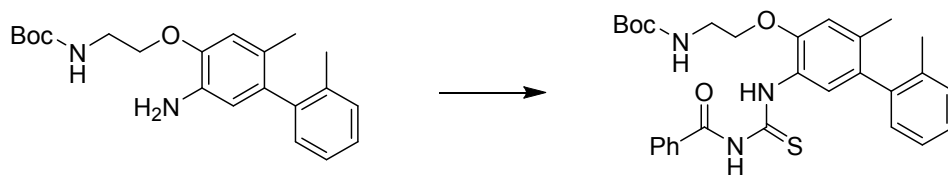
Synthesis of *tert*-butyl *N*-[2-({5-amino-2,2'-dimethyl-[1,1'-biphenyl]-4-yl}oxy)ethyl]carbamate



tert-Butyl *N*-[2-({2,2'-dimethyl-5-nitro-[1,1'-biphenyl]-4-yl}oxy)ethyl]carbamate (651 mg, 1.685 mmol) was dissolved in EtOH (30 mL) and the mixture was hydrogenated using the H-cube with 10% Pd/C as the catalyst. The mixture was evaporated *in vacuo* to yield *tert*-butyl *N*-[2-({5-amino-2,2'-dimethyl-[1,1'-biphenyl]-4-yl}oxy)ethyl]carbamate (583 mg, 97%). ¹H NMR (DMSO-*d*₆): δ 7.26-7.19 (m, 3H), 7.00 (d, *J* = 6.5 Hz, 1H), 6.67 (s, 1H), 6.33 (s, 1H), 4.70 (br s, 2H), 4.09 (br s, 1H), 3.92-3.89 (m, 2H), 3.36-3.32 (m, 2H), 2.02 (s, 3H), 1.84 (s, 3H), 1.42 (s, 9H). LCMS (method 3): *m/z* 357.1 [M+H]⁺, RT = 0.97 min. Purity 95%.

Step d:

Synthesis of *tert*-butyl *N*-{2-[(2,2'-dimethyl-5-[(phenylformamido)methanethioyl]amino}-[1,1'-biphenyl]-4-yl)oxy]ethyl}carbamate

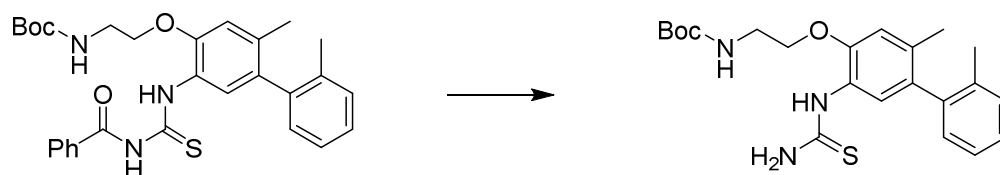


To a stirred solution of *tert*-butyl *N*-[2-({5-amino-2,2'-dimethyl-[1,1'-biphenyl]-4-yl}oxy)ethyl]carbamate (583 mg, 1.64 mmol) in DCM (10 mL) was added benzoyl

isothiocyanate (0.220 mL, 1.64 mmol). The reaction was stirred at ambient temperature. After 3 hours, the solvent was evaporated *in vacuo* to afford *tert*-butyl *N*-{2-[(2,2'-dimethyl-5-[[[(phenylformamido)methanethioyl]amino]-[1,1'-biphenyl]-4-yl]oxy]ethyl} carbamate as a yellow solid (850 mg, 100%) which was used without further purification. ¹H NMR (DMSO-*d*₆): δ 13.31 (s, 1H), 11.52 (s, 1H), 8.62 (s, 1H), 8.03-8.01 (m, 2H), 7.68-7.64 (m, 2H), 7.57-7.51 (m, 2H), 7.31-7.25 (m, 3H), 7.12-7.09 (m, 2H), 4.17 (t, *J* = 5.3 Hz, 2H), 3.47 (br s, 2H), 2.07 (s, 3H), 2.03 (s, 3H), 1.38 (s, 9H). LCMS (method 3): *m/z* 520.2 [M+H]⁺, RT = 1.49 min. Purity 90%.

Step e:

Synthesis of *tert*-butyl *N*-(2-{[5-(carbamothioylamino)-2,2'-dimethyl-[1,1'-biphenyl]-4-yl]oxy}ethyl)carbamate

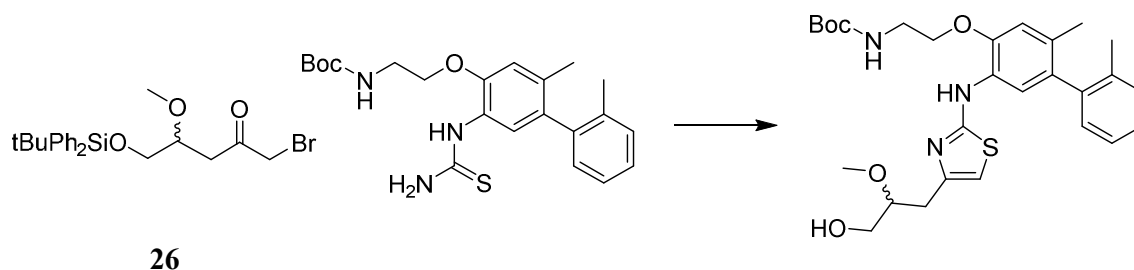


To a solution of *tert*-butyl *N*-{2-[(2,2'-dimethyl-5-[[[(phenylformamido)methanethioyl]amino]-[1,1'-biphenyl]-4-yl]oxy]ethyl} carbamate (850 mg, 1.64 mmol) in EtOH (50 mL) was added 1 M NaOH (6.54 mL, 6.54 mmol). The reaction was stirred at ambient temperature for 16 hours before the solvent was evaporated *in vacuo*. The residue was taken up in EtOAc (75 mL) and sat. NaHCO₃ (75 mL, aq.). The mixture was stirred for one hour and then the organic layer was separated and dried over MgSO₄. After filtration, the solvent was evaporated *in vacuo* to afford *tert*-butyl *N*-(2-{[5-(carbamothioylamino)-2,2'-dimethyl-[1,1'-biphenyl]-4-yl]oxy}ethyl)carbamate as a gray-

white solid (498 mg, 73%). ^1H NMR (DMSO- d_6): δ 8.95 (s, 1H), 7.60 (s, 1H), 7.45 (br s, 1H), 7.28-7.22 (m, 4H), 7.08-7.06 (m, 2H), 6.98 (s, 1H), 4.05 (t, J = 5.5 Hz, 2H), 3.40-3.36 (m, 2H), 2.06 (s, 3H), 1.98 (s, 3H), 1.42 (s, 9H). LCMS (method 3): m/z 416.1 $[\text{M}+\text{H}]^+$, RT = 1.14 min. Purity >90%.

Steps f and g:

Synthesis of *tert*-butyl *N*-{2-[(5-{[4-(3-hydroxy-2-methoxypropyl)-1,3-thiazol-2-yl]amino}-2,2'-dimethyl-[1,1'-biphenyl]-4-yl)oxy]ethyl}carbamate



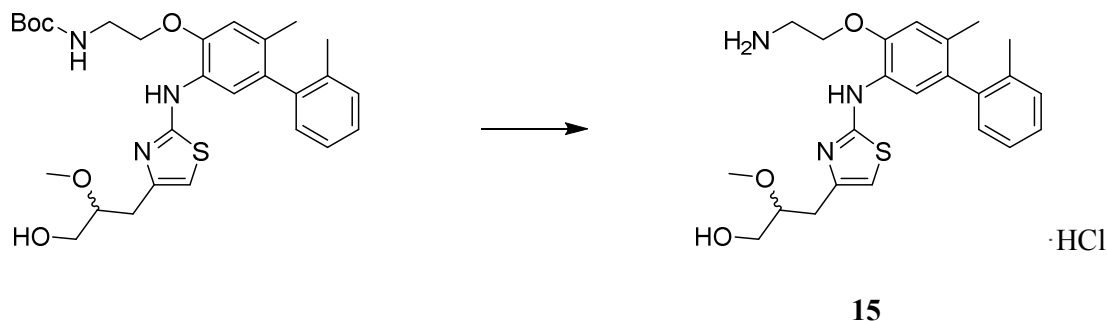
Preparation of intermediate 1-bromo-5-[(*tert*-butyldiphenylsilyl)oxy]-4-methoxypentan-2-one (**26**) is describe within the synthesis of 2-methoxy-3-[2-({4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-yl} amino)-1,3-thiazol-4-yl]propan-1-ol (**14**) as steps a-f.

To a stirred solution of 1-bromo-5-((*tert*-butyldiphenylsilyl)oxy)-4-methoxypentan-2-one (**26**) (200 mg, 0.445 mmol) in THF (10 mL) was added *tert*-butyl *N*-(2-{[5-(carbamothioylamino)-2,2'-dimethyl-[1,1'-biphenyl]-4-yl]oxy}ethyl)carbamate (185 mg, 0.445 mmol). The reaction mixture was heated at 61 °C for 3 hours and allowed to cool to ambient temperature. The solvent was evaporated *in vacuo* to afford *tert*-butyl *N*-[2-({5-[(4-{3-[(*tert*-butyldiphenylsilyl)oxy]-2-methoxypropyl}-1,3-thiazol-2-yl)amino]-2,2'-dimethyl-

[1,1'-biphenyl]-4-yl}oxy)ethyl]carbamate (345 mg) and was used without further purification. LCMS (method 3): m/z 766 $[M+H]^+$, $RT = 1.73$ min. Purity 85%. To a stirred solution of *tert*-butyl *N*-[2-(5-[(4-{3-[(*tert*-butyldiphenylsilyl)oxy]-2-methoxypropyl)-1,3-thiazol-2-yl]amino]-2,2'-dimethyl-[1,1'-biphenyl]-4-yl}oxy)ethyl]carbamate (345 mg, 0.450 mmol) in THF (2 mL) was added TBAF (1.35 mL, 1.35 mmol, 1 M in THF) at ambient temperature. The mixture was stirred for 3 hours. The reaction was evaporated *in vacuo* and the residue dissolved in DCM (10 mL). The organic layer was washed with water (10 mL) and evaporated *in vacuo*. The product was purified by flash column chromatography to yield *tert*-butyl *N*-{2-[(5-{[4-(3-hydroxy-2-methoxypropyl)-1,3-thiazol-2-yl]amino}-2,2'-dimethyl-[1,1'-biphenyl]-4-yl)oxy]ethyl}carbamate (179 mg, 75%). 1H NMR ($DMSO-d_6$): δ 9.07 (s, 1H), 8.00 (s, 1H), 7.30-7.22 (m, 3H), 7.08 (br s, 1H), 6.92 (s, 1H), 6.45 (s, 1H), 4.51 (br s, 1H), 4.10-4.02 (m, 4H), 3.43-3.34 (m, 3H), 3.15 (s, 3H), 2.62 (br s, 2H), 2.07 (s, 3H), 1.97 (s, 3H), 1.43 (s, 9H). LCMS (method 3): m/z 528.2 $[M+H]^+$, $RT = 1.09$ min. Purity >95%.

Step h:

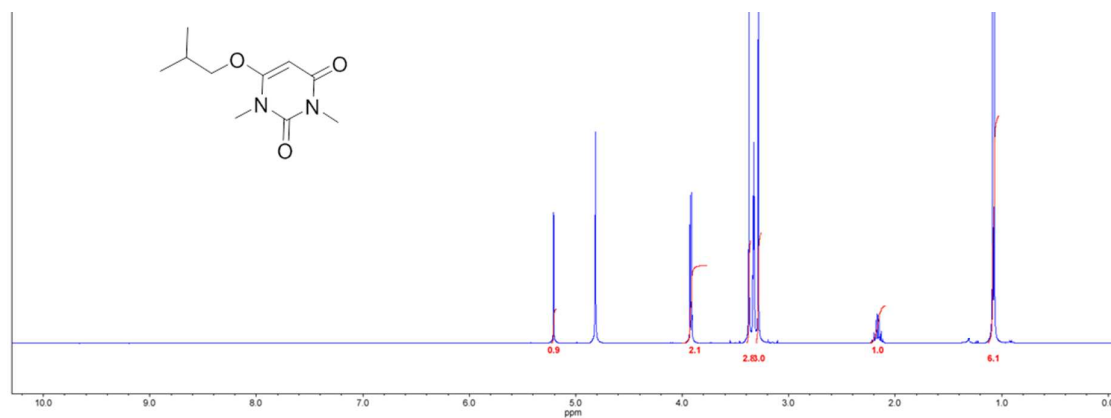
Synthesis of 3-(2-{[4-(2-aminoethoxy)-2',6-dimethyl-[1,1'-biphenyl]-3-yl]amino}-1,3-thiazol-4-yl)-2-methoxypropan-1-ol hydrochloride (**15**)



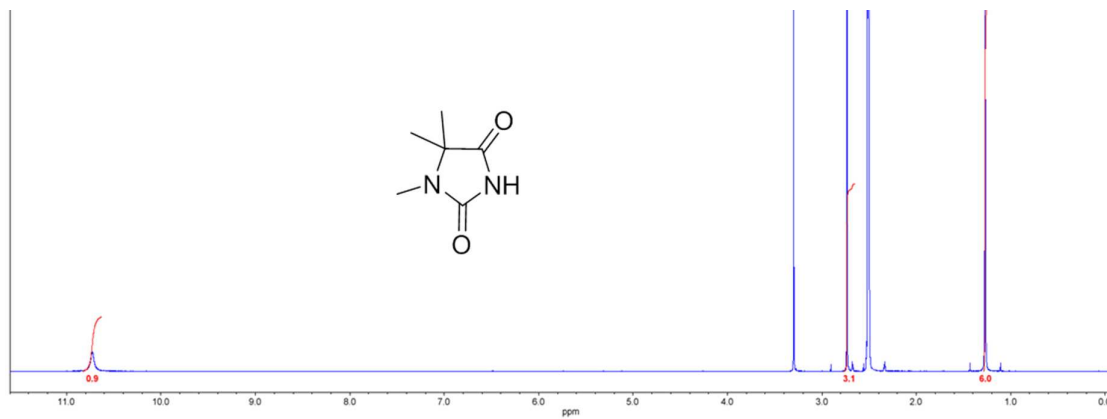
To a stirred solution of *tert*-butyl *N*-{2-[(5-{[4-(3-hydroxy-2-methoxypropyl)-1,3-thiazol-2-yl]amino}-2,2'-dimethyl-[1,1'-biphenyl]-4-yl)oxy]ethyl}carbamate (169 mg, 0.320 mmol) in 1,4-dioxane (2 mL) in air at ambient temperature was added a solution of HCl (4 mL, 16.0 mmol, 4 M in 1,4-dioxane) dropwise over 1 minute. The reaction mixture was stirred for 1 hour and then the solvent was evaporated *in vacuo* to yield 3-(2-{[4-(2-aminoethoxy)-2',6-dimethyl-[1,1'-biphenyl]-3-yl]amino}-1,3-thiazol-4-yl)-2-methoxypropan-1-ol hydrochloride (**15**) (141 mg, 95%). ¹H NMR (DMSO-*d*₆): δ 10.02 (br s, 1H), 8.37 (s, 3H), 7.85 (s, 1H), 7.31-7.22 (m, 3H), 7.10-7.08 (m, 1H), 7.02 (s, 1H), 6.54 (s, 1H), 4.29 (s, 2H), 3.44-3.33 (m, 5H), 3.19 (s, 3H), 2.72-2.61 (m, 2H), 2.07 (s, 3H), 2.00 (s, 3H). LCMS (method 3): *m/z* 428.1 [M+H]⁺, RT = 0.83 min. Purity >90%. HRMS: Found 428.2010, C₂₃H₂₉N₃O₃S [M+H]⁺ requires 428.2003 (Δ = 1.62 ppm). X-ray crystal structure gained in Lp-PLA₂.

14. ^1H NMR Spectra for Compounds 2-16

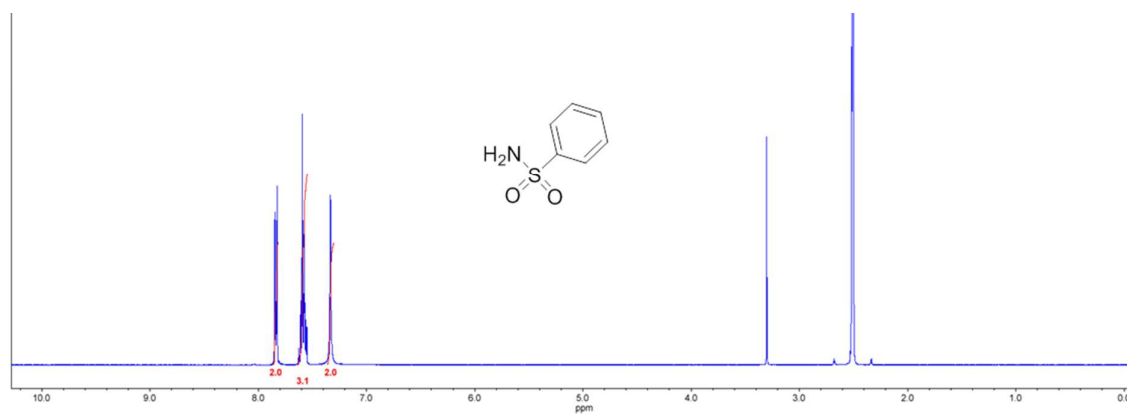
^1H NMR (CD_3OD) of 1,3-dimethyl-6-(2-methylpropoxy)-1,2,3,4-tetrahydropyrimidine-2,4-dione (**2**)



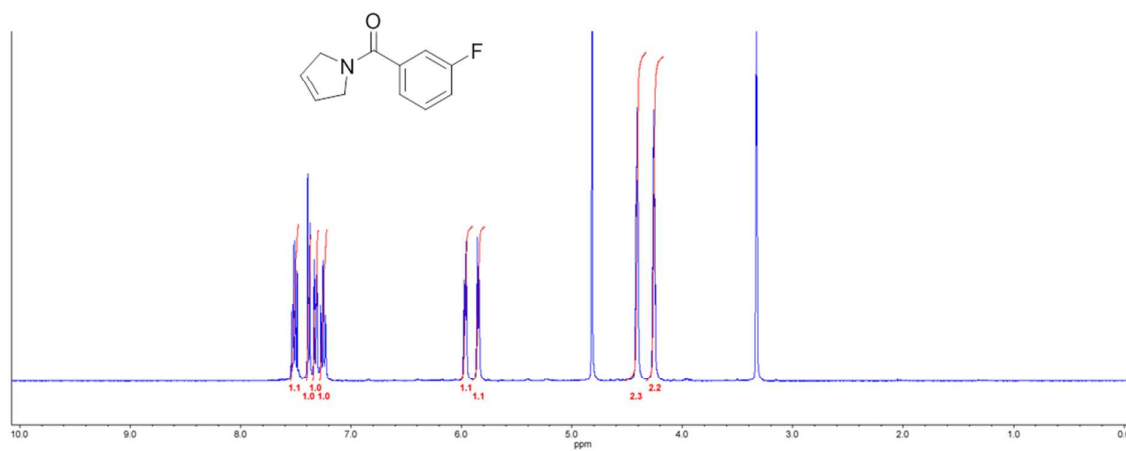
^1H NMR ($\text{DMSO}-d_6$) of 5,5-dimethylimidazolidine-2,4-dione (**3**)



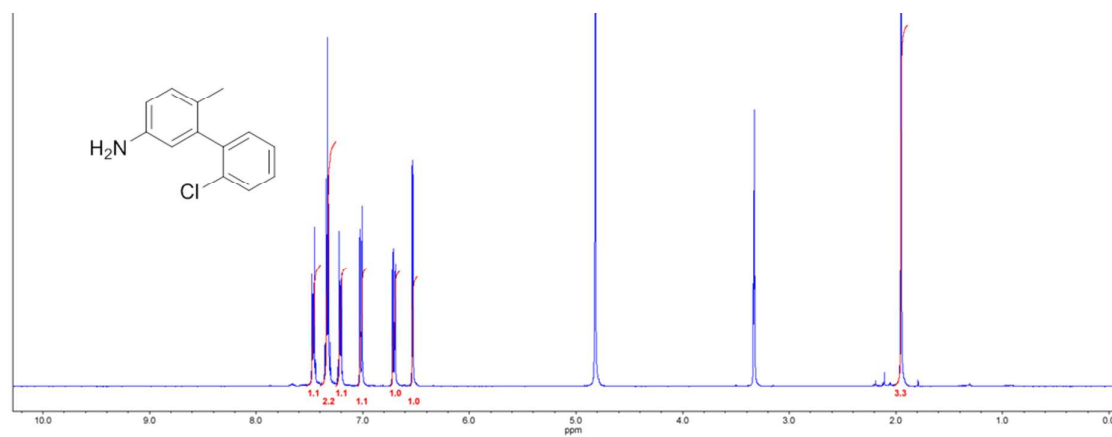
^1H NMR ($\text{DMSO}-d_6$) of benzenesulfonamide (**4**)



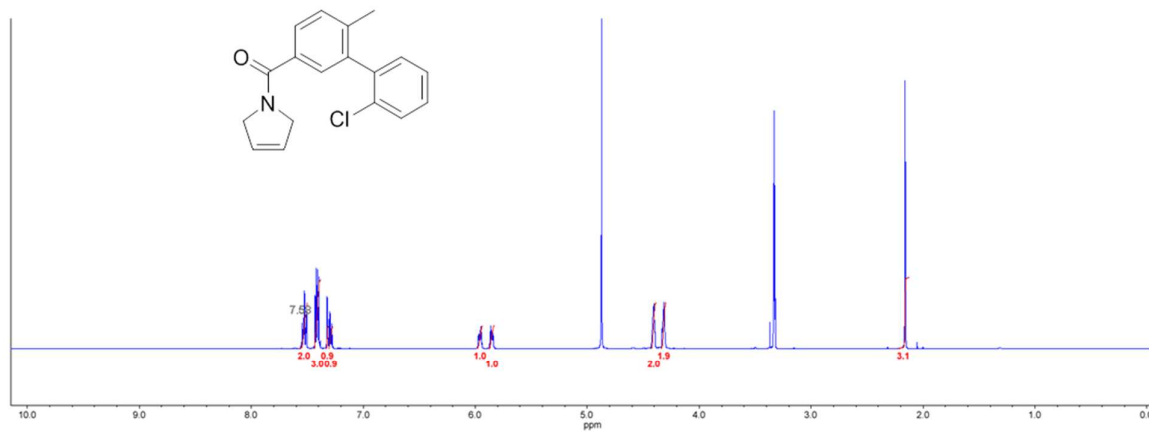
^1H NMR (CD_3OD) of 1-(3-fluorobenzoyl)-2,5-dihydro-1H-pyrrole (**5**)



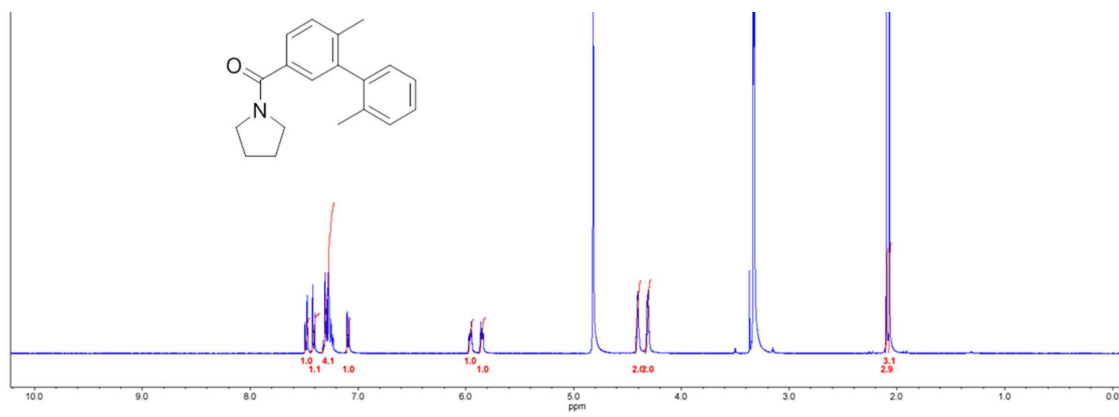
^1H NMR (CD_3OD) of 2'-chloro-6-methyl-[1,1'-biphenyl]-3-amine (**6**)



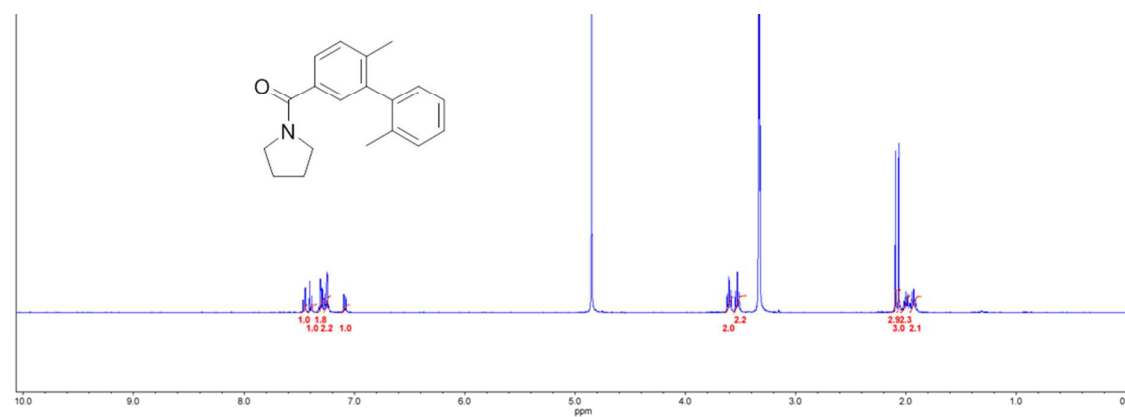
^1H NMR (CD_3OD) of 1-{2'-chloro-6-methyl-[1,1'-biphenyl]-3-carbonyl}-2,5-dihydro-1H-pyrrole (**7**)



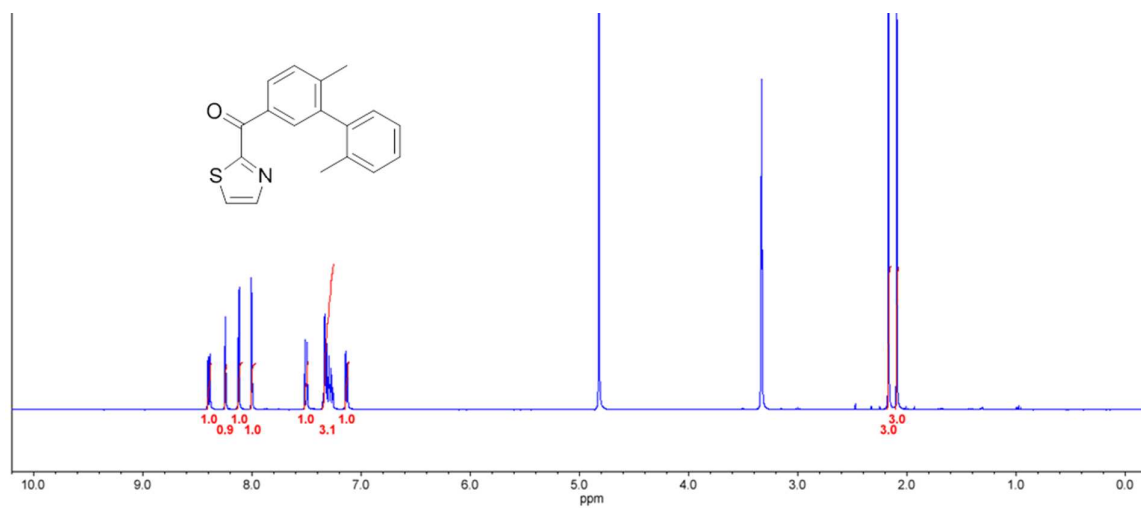
^1H NMR (CD_3OD) of 1-{2',6-dimethyl-[1,1'-biphenyl]-3-carbonyl}-2,5-dihydro-1H-pyrrole
(8)



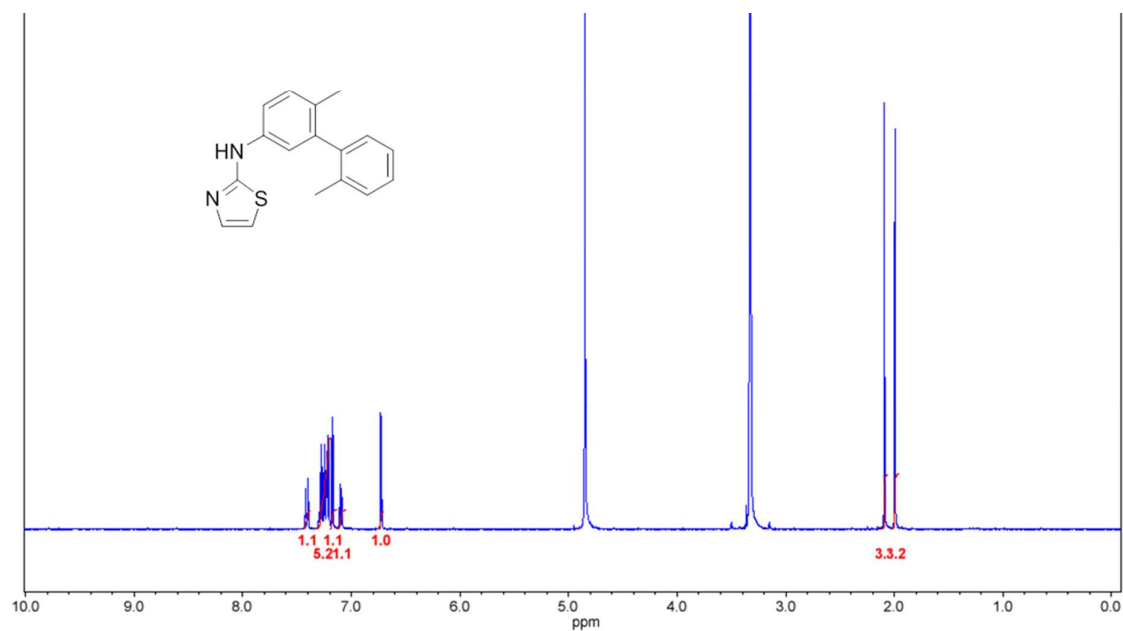
^1H NMR (CD_3OD) of 1-{2',6-dimethyl-[1,1'-biphenyl]-3-carbonyl}pyrrolidine (9)



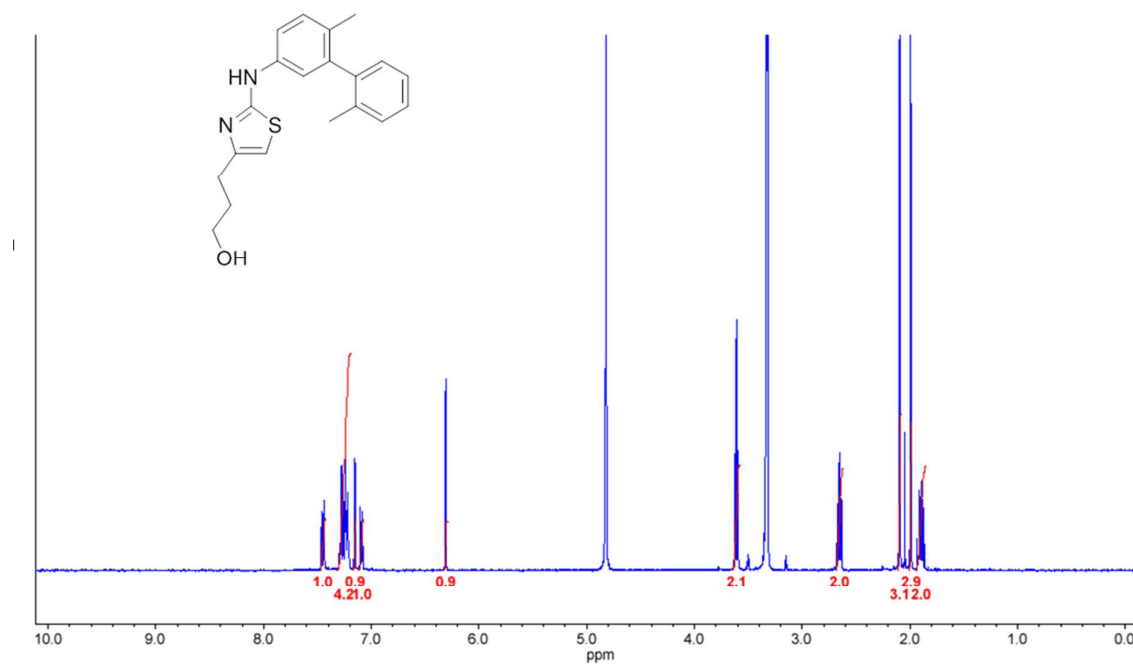
^1H NMR (CD_3OD) of 2-{2',6-dimethyl-[1,1'-biphenyl]-3-carbonyl}-1,3-thiazole (**10**)



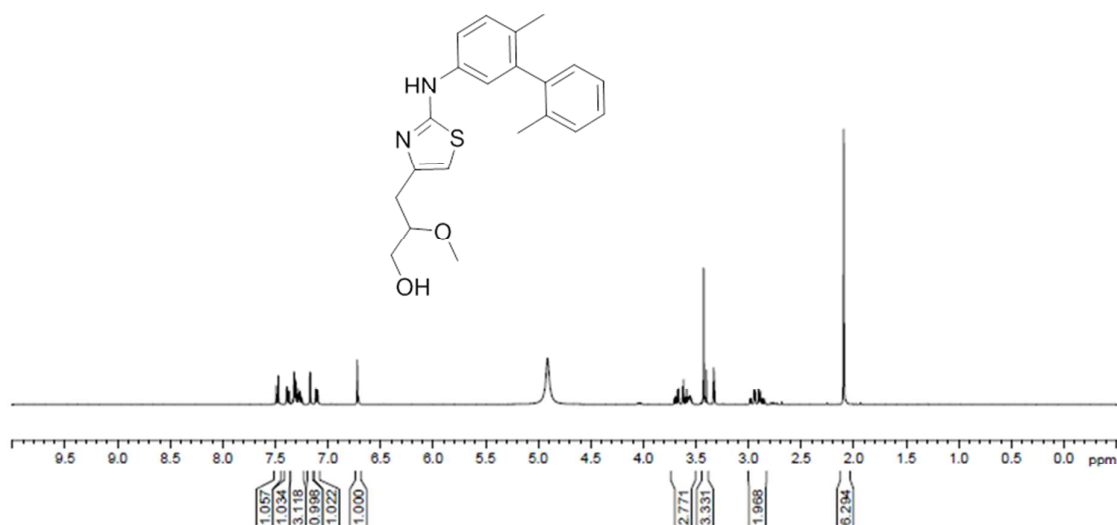
^1H NMR (CD_3OD) of *N*-{2',6-dimethyl-[1,1'-biphenyl]-3-yl}-1,3-thiazol-2-amine (**11**)



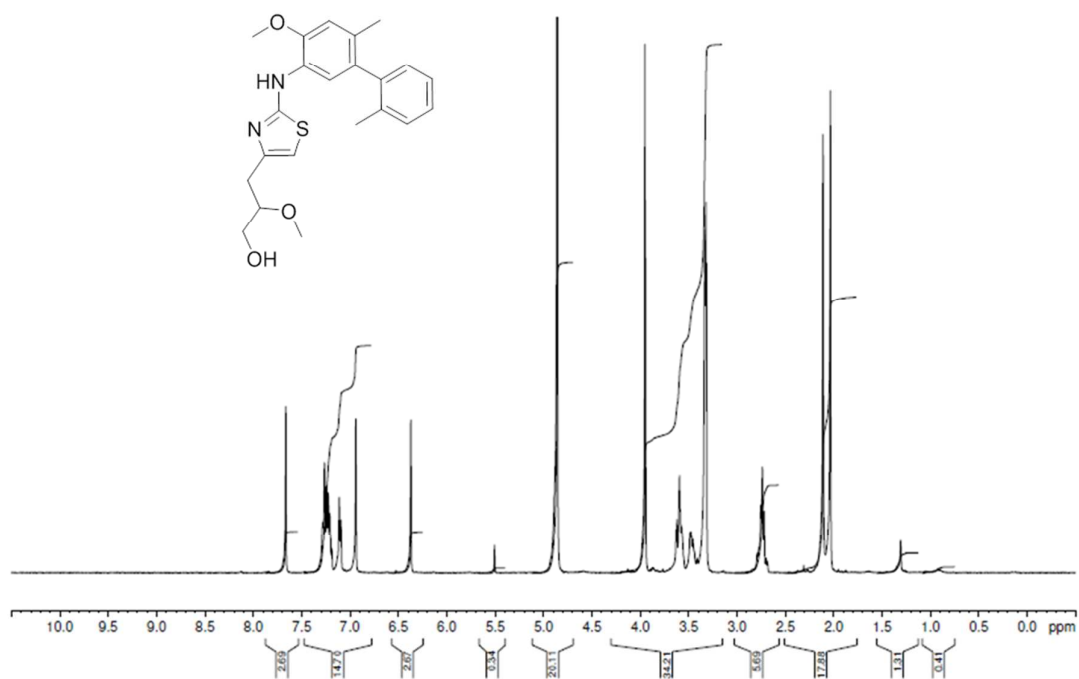
^1H NMR (CD_3OD) of 3-[2-(2',6-dimethyl-[1,1'-biphenyl]-3-yl)amino)-1,3-thiazol-4-yl]propan-1-ol (**12**)



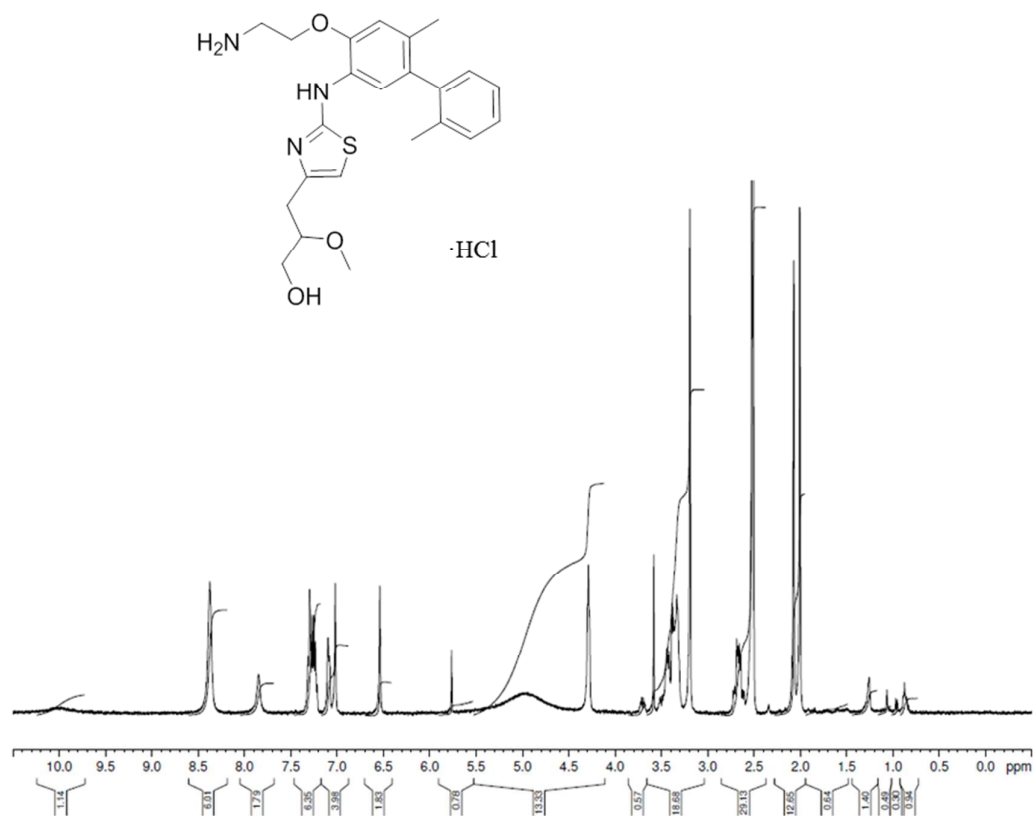
^1H NMR (CD_3OD) of 3-[2-(2',6-dimethyl-[1,1'-biphenyl]-3-yl)amino)-1,3-thiazol-4-yl]-2-methoxypropan-1-ol (**13**)



^1H NMR (CD_3OD) of 2-methoxy-3-[2-(4-methoxy-2',6-dimethyl-[1,1'-biphenyl]-3-yl)amino)-1,3-thiazol-4-yl]propan-1-ol (**14**)



^1H NMR ($\text{DMSO}-d_6$) of 3-(2-{[4-(2-aminoethoxy)-2',6-dimethyl-[1,1'-biphenyl]-3-yl]amino}-1,3-thiazol-4-yl)-2-methoxypropan-1-ol hydrochloride (**15**)



^1H NMR (CDCl_3) of 3-(3-{[4-(2-aminoethoxy)-2',6-dimethyl-[1,1'-biphenyl]-3-yl]amino}-1H-pyrazol-1-yl)-2-methoxypropan-1-ol trifluoroacetic acid (**16**)

