Supporting Information

Synthesis and characterization of a click-assembled 18-atom macrocycle that displays selective AXL kinase inhibitory activity

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1. COMPARISON OF NMR SPECTRA OF COMPOUNDS 1, 9 AND 7

$^1$H-NMR spectra of compounds 7, 9 and 1 are shown in Figure S1. In the three cases, an extremely broad doublet is observed from 4 to 5.2 ppm (highlighted by a frame). This flat-looking signal is indicative of numerous conformers coexisting at ambient temperature.

![NMR spectra comparison](image)

**Figure S1.** Comparison between the $^1$H-NMR spectra of macrocyclic derivatives 1, 9 and 7.
2. MACROCYCLE CONFORMATIONAL SAMPLING

2.1. Macrocycle sampling of compound 1

The initial structure of compound 1 was constructed in Maestro and minimized to a local minimum with MacroModel using an OPLS 2005 force field in a steepest descent method for a maximum of 200 iterations, and a convergence threshold of 0.05 in water. The final structure was next submitted to a macrocycle conformational sampling with MacroModel and the following parameters: 5000 quench cycles of an stochastic molecular dynamics calculation at 1000 K followed by another 0.5 ps at 300 K, and 5000 large-scale low-mode (LLMOD) search steps. Eigenvectors were determined for each global minimum and the planar torsion sampling options enhanced. An OPLS_2005 force field was used for the minimization with a GB/SA (water) electrostatic treatment, with an energy window of 10 kcal/mol, and an RMSD cutoff of 0.75.

Macrocycle 1 sampling with MacroModel yielded 155 different conformational structures, which were saved as independent PDB files. They are classified into 4 groups depending on the spatial disposition of its benzene ring in relation to the pyrazolopyrimidine heterocyclic moiety. Below is a table containing the potential energy and simply root-mean-square deviation (RMSD) values of all the conformers.

**Table S1. Potential Energy (kJ/mol) and RMSD of 1 conformers (ocl) obtained after macrocycle sampling.**

| Conformer | Pot. Energy (kJ/mol) | RMSD |
|-----------|----------------------|------|
| ocl_1     | -844.751             | 0    |
| ocl_2     | -844.570             | 1.172|
| ocl_3     | -844.535             | 0.972|
| ocl_4     | -844.516             | 0.778|
| ocl_5     | -844.498             | 1.135|
| ocl_6     | -844.409             | 3.513|
| ocl_7     | -844.180             | 1.156|
| ocl_8     | -843.872             | 1.197|
| ocl_9     | -843.735             | 1.589|
| ocl_10    | -843.530             | 1.378|
| ocl_11    | -843.422             | 1.151|
| ocl_12    | -843.411             | 1.395|
| ocl_13    | -842.880             | 1.293|
| ocl_14    | -841.694             | 1.365|
| ocl_15    | -841.615             | 1.248|
| ocl_16    | -841.504             | 1.088|
| ocl_17    | -841.500             | 1.253|
| ocl_18    | -840.726             | 1.087|
| ocl_19    | -840.647             | 1.601|
| ocl_20    | -840.279             | 1.460|
| ocl_21    | -840.233             | 0.921|
| ocl_22    | -840.190             | 1.098|
| ocl_23    | -840.190             | 1.153|
| ocl_24    | -839.866             | 1.344|
| ocl_25 | -839.584 | 1.071 |
| ocl_26 | -839.571 | 1.084 |
| ocl_27 | -839.262 | 1.359 |
| ocl_28 | -839.194 | 1.090 |
| ocl_29 | -839.121 | 1.355 |
| ocl_30 | -838.850 | 1.546 |
| ocl_31 | -838.658 | 0.805 |
| ocl_32 | -838.562 | 1.402 |
| ocl_33 | -838.557 | 1.558 |
| ocl_34 | -838.225 | 1.664 |
| ocl_35 | -838.109 | 1.284 |
| ocl_36 | -837.898 | 1.416 |
| ocl_37 | -837.820 | 1.269 |
| ocl_38 | -837.733 | 1.166 |
| ocl_39 | -837.581 | 1.505 |
| ocl_40 | -837.555 | 1.357 |
| ocl_41 | -837.553 | 1.761 |
| ocl_42 | -837.309 | 3.091 |
| ocl_43 | -837.282 | 3.588 |
| ocl_44 | -837.156 | 3.318 |
| ocl_45 | -836.852 | 1.604 |
| ocl_46 | -836.215 | 1.578 |
| ocl_47 | -835.961 | 1.293 |
| ocl_48 | -835.371 | 1.210 |
| ocl_49 | -834.983 | 3.417 |
| ocl_50 | -834.069 | 1.147 |
| ocl_51 | -833.834 | 3.451 |
| ocl_52 | -833.821 | 1.261 |
| ocl_53 | -833.668 | 1.576 |
| ocl_54 | -833.129 | 2.916 |
| ocl_55 | -830.643 | 1.505 |
| ocl_56 | -830.537 | 1.543 |
| ocl_57 | -830.374 | 1.414 |
| ocl_58 | -830.341 | 1.586 |
| ocl_59 | -829.414 | 1.376 |
| ocl_60 | -826.756 | 1.586 |
| ocl_61 | -826.164 | 3.299 |
| ocl_62 | -826.075 | 0.956 |
| ocl_63 | -825.968 | 1.258 |
| ocl_64 | -825.878 | 3.488 |
| ocl_65 | -825.754 | 3.486 |
| ocl_66 | -825.392 | 1.204 |
| ocl_67 | -825.377 | 1.174 |
| ocl_68 | -825.148 | 1.523 |
| ocl_69 | -824.990 | 1.153 |
| ocl_70 | -824.202 | 1.306 |
| ocl_71 | -823.531 | 1.133 |
| ocl_72 | -823.459 | 3.427 |
| ocl_73 | -823.471 | 1.234 |
| ocl_74 | -823.444 | 2.579 |
| ocl_75 | -823.428 | 0.907 |
| ocl_76 | -823.362 | 2.015 |
| ocl_77 | -823.197 | 1.270 |
| ocl_78 | -823.055 | 2.474 |
| ocl_79 | -822.932 | 1.332 |
| ocl_80 | -822.906 | 1.319 |
| ocl_81 | -822.768 | 1.428 |
| ocl_82 | -822.364 | 1.343 |
| ocl_83 | -822.277 | 1.315 |
| ocl_84 | -822.181 | 1.143 |
| ocl_85 | -822.015 | 3.691 |
| ocl_86 | -821.629 | 1.123 |
| ocl_87 | -821.457 | 1.609 |
| ocl_88 | -821.427 | 1.406 |
| ocl_89 | -821.048 | 1.492 |
| ocl_90 | -820.770 | 1.293 |
| ocl_91 | -820.703 | 1.166 |
| ocl_92 | -820.663 | 1.022 |
| ocl_93 | -820.629 | 1.219 |
| ocl_94 | -820.616 | 1.167 |
| ocl_95 | -820.580 | 1.165 |
| ocl_96 | -820.513 | 1.251 |
| ocl_97 | -820.493 | 3.506 |
| ocl_98 | -820.279 | 0.862 |
| ocl_99 | -819.955 | 2.187 |
| ocl_100 | -819.739 | 2.461 |
| ocl_101 | -819.677 | 2.585 |
| ocl_102 | -819.520 | 1.368 |
| ocl_103 | -819.422 | 1.613 |
| ocl_104 | -819.351 | 1.375 |
| ocl_105 | -819.328 | 2.820 |
| ocl_106 | -819.183 | 3.358 |
| ocl_107 | -819.046 | 2.717 |
| ocl_108 | -819.025 | 1.710 |
| ocl_109 | -818.929 | 1.712 |
| ocl_110 | -818.796 | 1.087 |
| ocl_111 | -818.598 | 2.142 |
| ocl_112 | -818.403 | 1.881 |
| ocl_113 | -818.013 | 1.487 |
| ocl_114 | -817.643 | 2.172 |
| ocl_115 | -817.643 | 3.427 |
| ocl_116 | -817.599 | 1.648 |
| ocl_117 | -817.068 | 2.059 |
| ocl_118 | -816.706 | 2.952 |
| ocl_119 | -816.616 | 1.260 |
| ocl_120 | -816.602 | 1.272 |
| ocl_121 | -816.376 | 2.613 |
| ocl_122 | -816.093 | 1.407 |
| ocl_123 | -816.058 | 1.227 |
| ocl_124 | -815.872 | 1.865 |
| ocl_125 | -815.535 | 1.121 |
| ocl_126 | -815.447 | 1.113 |
| ocl_127 | -815.373 | 3.604 |
| ocl_128 | -815.342 | 2.506 |
| ocl_129 | -815.212 | 2.204 |
| ocl_130 | -815.041 | 2.005 |
| ocl_131 | -815.027 | 1.527 |
| ocl_132 | -814.753 | 1.428 |
| ocl_133 | -814.540 | 1.366 |
| ocl_134 | -814.482 | 2.667 |
| ocl_135 | -813.778 | 1.727 |
| ocl_136 | -813.735 | 1.468 |
| ocl_137 | -812.181 | 1.584 |
| ocl_138 | -811.477 | 1.268 |
| ocl_139 | -811.145 | 2.528 |
| ocl_140 | -811.110 | 2.549 |
| ocl_141 | -810.829 | 2.675 |
| ocl_142 | -809.943 | 3.394 |
| ocl_143 | -809.723 | 2.220 |
| ocl_144 | -809.420 | 2.550 |
| ocl_145 | -809.320 | 2.408 |
| ocl_146 | -809.066 | 2.063 |
| ocl_147 | -806.772 | 1.739 |
| ocl_148 | -806.494 | 2.511 |
| ocl_149 | -806.381 | 2.350 |
| ocl_150 | -805.110 | 2.481 |
| ocl_151 | -804.940 | 2.435 |
| ocl_152 | -804.823 | 2.505 |
| ocl_153 | -803.942 | 2.718 |
2.2. Results from macrocycle sampling of 7YS

The 3D structure of native ligand 7YS was obtained from the Protein Data Bank web page (PDB ID: 5U6B) and submitted to a macrocycle conformational sampling with MacroModel as described above for 1. Fifteen different conformational structures were obtained (see Figure S2). The obtained conformational structures of 7YS were saved as independent PDB files, together with its native conformation.

Figure S2. Superimposed 3D-structure model for the 15 7YS conformers. For clarity, the conformers have been superimposed through its pyrazine ring.

A table containing the potential energy and RMSD values of all the conformers found for macrocyclic ligand 7YS is shown below.

Table S2. Potential Energy (kJ/mol) and RMSD of 7YS conformers obtained after macrocycle sampling.

| Conformer | Pot. Energy (kJ/mol) | RMSD  |
|-----------|----------------------|-------|
| 7YS_1     | 21.561               | 0     |
| 7YS_2     | 21.894               | 0.882 |
| 7YS_3     | 29.810               | 0.316 |
| 7YS_4     | 35.358               | 1.297 |
| 7YS_5     | 35.566               | 1.652 |
| 7YS_6     | 36.257               | 0.845 |
| 7YS_7     | 41.859               | 1.632 |
| 7YS_8     | 42.323               | 1.266 |
| 7YS_9     | 48.405               | 1.431 |
| 7YS_10    | 49.091               | 1.007 |
| 7YS_11    | 51.324               | 0.979 |
| 7YS_12    | 57.884               | 2.599 |
| 7YS_13    | 57.912               | 0.973 |
| 7YS_14    | 59.245               | 2.563 |
| 7YS_15    | 60.490               | 2.450 |
2.3 Cluster analysis of the 155 macrocycle sampling conformers of compound 1

A cluster analysis of an ensemble of the 155 conformers was carried out to determine the conformer representatives. This analysis was achieved through an implementation of the method described by Kelley et al\textsuperscript{3} on UCSF Chimera.\textsuperscript{4} A total of 14 clusters were obtained been clusters 1, 2 and 3 the most populated with 41, 37 and 26 conformers, respectively (Figure S3 and Table S3).

![Figure S3](image)

**Figure S3.** (A) 3D structure models (superimposed through its pyrazolopyrimidine ring) of all the 155 conformers of the 14 clusters of compound 1. (B) Representative conformers (superimposed through its pyrazolopyrimidine ring) for the first 7 clusters of compound 1. Cluster 1 (yellow), cluster 2 (blue), cluster 3 (purple), cluster 4 (dark green), cluster 5 (aubergine), cluster 6 (green), cluster 7 (pink).

**Table S3.** Energy, energy differences, cluster population and representative conformer of the 14 clusters of compound 1.

| # cluster | # total conf. | Representative conf. | Pot. Energy (kJ/mol) | RMSD | RMSD (macrocycle atoms) |
|-----------|---------------|----------------------|----------------------|------|-------------------------|
| 1         | 41            | comp1_94             | -820.616             | 0    | 0                       |
| 2         | 37            | comp1_104            | -819.351             | 1.025| 1.213                   |
| 3         | 26            | comp1_08             | -843.872             | 2.102| 2.133                   |
| 4         | 10            | comp1_107            | -819.046             | 4.358| 5.079                   |
| 5         | 9             | comp1_06             | -844.409             | 5.923| 6.916                   |
| 6         | 6             | comp1_118            | -816.706             | 5.719| 6.703                   |
| 7         | 5             | comp1_99             | -819.955             | 3.888| 4.456                   |
| 8         | 4             | comp1_145            | -809.320             | 5.046| 5.860                   |
| 9         | 4             | comp1_100            | -818.796             | 4.651| 5.343                   |
As shown in Figure S3B, the conformers included in cluster 1 (yellow) and 5 (grey), place its benzene ring in a *cis* disposition with respect to the pyrazolopyrimidine ring although from opposite sides of the bicycle. Conformers comprising clusters 2 (blue), 3 (purple) and 6 (light green) place its benzene ring in a *trans* disposition, with cluster 6 facing the opposite side of the bicyclic ring. Cluster 4 (dark green) and 7 (pink), on the other hand, show a stretched conformation of the macrocyclic ring, as do other minority conformers from clusters 8 to 14. The high number of possible clusters and conformers in compound 1 could be attributed to the marked flexibility of the newly added CH$_2$ group and triazole ring at the C6 position of the pyrazolopyrimidine ring. This feature is better appreciated in Figure S3B where the CH$_2$ group is oriented in several dispositions and could be supported by the experimental behavior of its protons in $^1$H-NMR, suggesting a rapid conformational exchange between conformers at room temperature (see manuscript).

3. DOCKING STUDIES

3.1. Protocol

Docking studies were carried out with Autodock 4.2.6 (AD4)\(^1\) on the crystal structure of tyrosine kinase receptor AXL (PDB ID: 5U6B) and RET (PDB ID: 2IVV). Ligands PDB files were prepared for docking using the prepare_ligand4.py script included MGLTools 1.5.4.,\(^2\) and protein structures using Maestro Protein Preparation wizard.\(^3\) Water and ligand molecules were removed and charges and non-polar hydrogen atoms were added at pH 7.0. The produced structures were saved as PDB files and prepared for docking using the prepare_receptor4.py script from MGLTools. AD4 was used to automatically dock the ligands into the ATP binding pocket of the prepared protein structures. A docking grid was set with the following grid parameters: 61 Å × 61 Å × 61 Å with 0.375 Å spacing, and centered on the ATP binding pocket. AD4 parameter file was set to 100 GA runs, 2,500,000 energy evaluations and a population size of 150. The Lamarckian genetic algorithm local search (GALS) method was used for the docking calculations. All dockings were performed with a population size of 250 and a Solis and Wets local search of 300 rounds was applied with a probability of 0.06. A mutation rate of 0.02 and a crossover rate of 0.8 were used. The docking results from each of the 100 calculations were clustered based on root-mean square deviation (RMSD) (solutions differing by less than 2.0 Å) between the Cartesian coordinates of the atoms and were ranked on the basis of free energy of binding.

3.2. Docking of ligand 7YS

Receptor AXL is one of the three members of the tyrosine kinase receptors family known as TAM, also including TYRO3 and MER. A crystal structure of AXL has been recently published in which its kinase domain is in complex with a macrocyclic inhibitor\(^6\) in two conformational states, one of them meeting most of the known structural constraints for an active kinase domain (PDB ID: 5U6B).\(^7\) For the docking
studies on AXL, the structure of the active conformation was selected (PDB ID: 5U6B, monomer B). Redocking of the cognate ligand 7YS showed a similar binding pose to that of the crystal complex with a RMSD of 0.126 Å (Figure S4A), validating the docking protocol. Next, cognate ligand 7YS was submitted to. The fifteen different conformational structures obtained from macrocycle sampling with MacroModel (see above) were docked on AXL active site. The resulting binding pose showed an almost identical binding mode with the cognate ligand 7YS (RMSD of 0.189 Å) (Figure S4B).

Figure S4. (A) Cognate ligand 7YS (light yellow) and re-docked lower energy pose of 7YS (light blue) on the active site of AXL (grey) (PDB ID: 5U6B). (B) Cognate ligand 7YS (light yellow) and best docked pose of 7YS (yellow) after macrocyclic sampling on the active site of AXL (grey) (PDB ID: 5U6B). DFG motif (cyan), hinge region (lime), G-loop (dark green), and H-bonds (magenta dotted lines). (C) Structure of 7YS.
3.3. Docking of macrocycle 1

Similarly, the 155 conformational structures of 1 were submitted to docking studies with AD4 in receptor AXL (PDB ID: 5U6B). Since the available crystal structure of the AXL kinase domain is in complex with macrocyclic ligand 7YS, the predicted binding poses of 1 and 7YS were compared. The resulting binding pose of 1, together with cognate ligand 7YS, are showed in Figure S5.

![Figure S5. (A) Cognate ligand 7YS (pink) and predicted binding pose of 1 (yellow) on the active site of AXL (grey) (PDB ID: 5U6B); (B) Cognate ligand 7YS (pink) and predicted binding pose of 1 (yellow) with coulombic representation of AXL (PDB ID: 5U6B). Red and blue areas represent negatively and positively polarized areas, respectively; (C) Front view of cognate ligand 7YS (pink) and predicted binding pose of 1 (yellow). DFG motif (cyan), hinge region (lime), G-loop (dark green), and H-bonds (magenta dotted lines).](image)

As seen in the figure, the pyrazine and benzene rings in 7YS and 1, respectively, are superimposed in a hydrophobic pocket at the ATP-binding site of AXL (Figure S5C). Towards the opening of the cleft, and near the hinge region, are located the pyrazole ring of 7YS and the N1-triazole of 1 (Figure S5A and C). At the opposite side, deep inside the ATP-binding cleft, and near the catalytically important Lys567, is placed the C6 triazole ring in 1. This allows the ligand to form two H-bonds with the lysine residue and forcing the cationic piperazine arm to orient outside the catalytic site, and towards a negatively charged region in the G-loop, forming a H-bond with Gly545 in its backbone chain. While this G-loop region is of flexible nature and the H-bond with this region might not be so easily reached, this binding pose of 1 is partly achieved thanks to the two H-bonds of the newly added C6 triazole.

Ligand eSM119 was also docked on AXL, adopting a different orientation when compared to the binding pose of 1. This open-chain ligand wraps itself inside the ATP-binding site and places its pyrazolopyrimidine ring on the hinge region, forming a H-bond through its methylamino group with the backbone chain (Figure S6). Its piperazine ring, on the other hand, faces the DFG motif forming a strong cationic H-bond with Asp690 (Figure S6).
Figure S6. Ligand eSM119 (light brown) and 1 (gold) docked on the active site of AXL (grey) (PDB ID: 5U6B). DFG motif (cyan), hinge region (lime), G-loop (dark green), and H-bonds (magenta dotted lines).

Next, docking studies with the 155 macrocyclic conformations of 1 were carried out in receptor tyrosine kinase RET (PDB ID: 2IVV). The X-ray structure of RET (PDB ID: 2IVV) was downloaded and superimposed on the structure of AXL (PDB ID: 5U6B) using Maestro. The co-crystal ligand of 2IVV and crystal structure of 2IVV were processed as described in the Material and Methods section and docked with AD4 on RET using the same docking parameters described previously for the docking of 7YS and 1 in AXL. Re-docking of the cognate ligand PP1 (see (PDB ID: 2IVV) showed a similar binding pose to that of the crystal complex validating the docking protocol on RET (results not shown).

The predicted binding mode of 1 in RET and AXL are presented in Figure S7. In both enzymes, ligand 1 virtually superimpose its macrocycle ring, placing the benzene ring in a hydrophobic pocket and displaying the C6-triazole toward the DFG motif. However, in RET (Figure S7A), the G-loop is displaced toward the DFG motif closing the cleft and forcing 1 to move upward, hence removing its C6-triazole from the proximity of Lys 758 (Figure S6A and B). As a result, the H-bonds with Lys758 cannot take place and only a single cationic H-bond is formed at the G-loop between the protonated piperazine ring and Glu732, yielding a lower affinity binding pose. This characteristic can also be seen when considering the coulombic surface of both enzymes (Figures S7C and D). The higher binding affinity pose of 1 in AXL is not allowed in RET due to steric clash. Instead, the ligand is slightly moved outside the cleft adopting the previously described lower affinity pose. These predicted results suggest a lower selectivity of compound 1 toward RET and could explain the higher activity observed experimentally in AXL, which can be attributed to the newly added C6-triazole allowing two new H-bonds with Lys567. As described above, this feature does not take place with RET kinase.
Figure S7. (A) Predicted binding pose of 1 in RET (light green) and AXL (yellow) on the active site of RET (orange) (PDB ID: 2IVV); (B) Predicted binding pose of 1 in RET (light green) and AXL (yellow) on the active site of AXL (grey) (PDB ID: 5U6B); (C) Coulombic representation of RET with predicted binding pose of 1 in RET (light green) and AXL (yellow); (D) Coulombic representation of AXL with predicted binding pose of 1 in RET (light green) and AXL (yellow). Red and blue areas represent negatively and positively polarized areas, respectively. DFG motif (cyan), hinge region (lime), G-loop (dark green), and H-bonds (magenta dotted lines).

When comparing the binding poses of 1 and eSM119 in AXL (Figure S8), the former appears to have a stronger interaction with the binding site than the later and therefore should possess a higher inhibitory activity. However, some characteristic must be taking into consideration for both poses to account for the experimental results. The binding pose of 1 (see Figures S6 and S7) comes from comp1_136, one of the less energetically-stable conformers obtained in the macrocycle sampling of 1 (see Table 1) and belonging to cluster 2 (Figure S3), which might not be so easily attained under the experimental conditions. Moreover, while 1 is able to form up to 3 stable H-bonds with the kinase active site, the one with the G-loop might not be so easily achieved due to the flexible nature of this region and the rigidity of the piperazino-pyrimidine arm. On the other hand, eSM119 is an open chain inhibitor and the binding pose can be, in principle, more energetically favorable. On top of that, eSM119 is forming two stable H-bonds with two stationary and conserved regions in the protein, the Hinge and the DFG motif, been the piperazine H-bond with Asp690 in the DFG motif one of the strongest because of its cationic nature.
To explain the lack of selectivity observed in compound eSM119 with both AXL and RET kinases, further docking studies of this compound on RET protein were carried out following the protocol described above. The binding poses of eSM119 on AXL and RET are shown on Figure S8.

**Figure S8.** (A) Predicted binding poses of eSM119 in AXL (light brown) and RET (pink) on the active site of RET kinase (orange) (PDB ID: 2IVV). (B) Predicted binding poses of eSM119 in AXL (light brown) and RET (pink) on the active site of AXL kinase (grey) (PDB ID: 5U6B). DFG motif (cyan), hinge region (lime), G-loop (dark green), and H-bonds (magenta dotted lines).

As Figure S8 shows, eSM119 bind similarly in both kinases forming a strong cationic H-bond with the important aspartic residue of the DFG motif. This feature might explain the lack of selectivity of eSM119 toward AXL and RET and its ability to inhibit both enzymes with similar efficacy. While eSM119 is able to bind both AXL and RET showing similar inhibitory activities, compound 1, on the contrary, is not able to establish any recognizable interaction with this residue but only with Lys567 in AXL kinase (Figure S6A and S6B). These results allow us to draw some insights of the way these inhibitors work. One of them is the high levels of inhibition achieved when the Aspartic residue of DFG is targeted and the ability of open-chain inhibitors, such as eSM119, to reach this residue. The second is the fact that by including rigidity in our inhibitors with the construction of cyclic compound 1, the residue Lys567 can be selectively targeted on AXL kinase allowing the inhibition of this enzyme.
4. DOSE RESPONSE STUDY OF COMPOUND 1 IN MV4-11 CELLS

MV4-11 cells were treated with compound 1 at a range of concentrations (0.3 to 30 μM). The resulting dose response curve is shown in Figure S8, which was used to calculate an EC50 value of 16.6 μM.

![Figure S8](image_url)

Cell Viability [%]

Log [1] (M)

Figure S9. Dose response curve of MV4-11 cells treated with compound 1. Error bars: ± SD from n = 2.

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4. NMR SPECTRA

4.1. $^1$H NMR of compound 4

Figure S10.
4.2. $^1$H and $^{13}$C NMR of compound 5

Figure S11.
4.3. $^1$H and $^{13}$C NMR of compound 7

Figure S12.
4.4. $^1$H and $^{13}$C NMR of compound 9

Figure S13.
4.5. $^1$H and $^{13}$C NMR of compound 1

Figure S14. $^1$H and $^{13}$C NMR of compound 1 at 27 °C.
Figure S15. $^1$H NMR of compound 1 at 80 °C.