SUPPORTING INFORMATION

for

Efficient synthesis of hydroxy-substituted 2-aminobenzo[*d*]thiazole-6-carboxylic acid derivatives as new building blocks in drug discovery

Martina Durcik,[a] Žan Toplak,[a] Nace Zidar,[a] Janez Ilaš,[a] Anamarija Zega,[a] Danijel Kikelj,[a] Lucija Peterlin Mašič[a] and Tihomir Tomašič[a,*]

[a]University of Ljubljana, Faculty of Pharmacy, Aškerčeva c. 7, SI-1000 Ljubljana, Slovenia

*Email: Tihomir Tomašič - Tihomir.Tomasic@ffa.uni-lj.si

Table of contents

S1. HPLC-MS analysis of cyclisation mechanism for compound 4b (5b).

Figure S1. HPLC-MS analysis of the reaction mixture before the addition of bromine (starting compound **4b**).

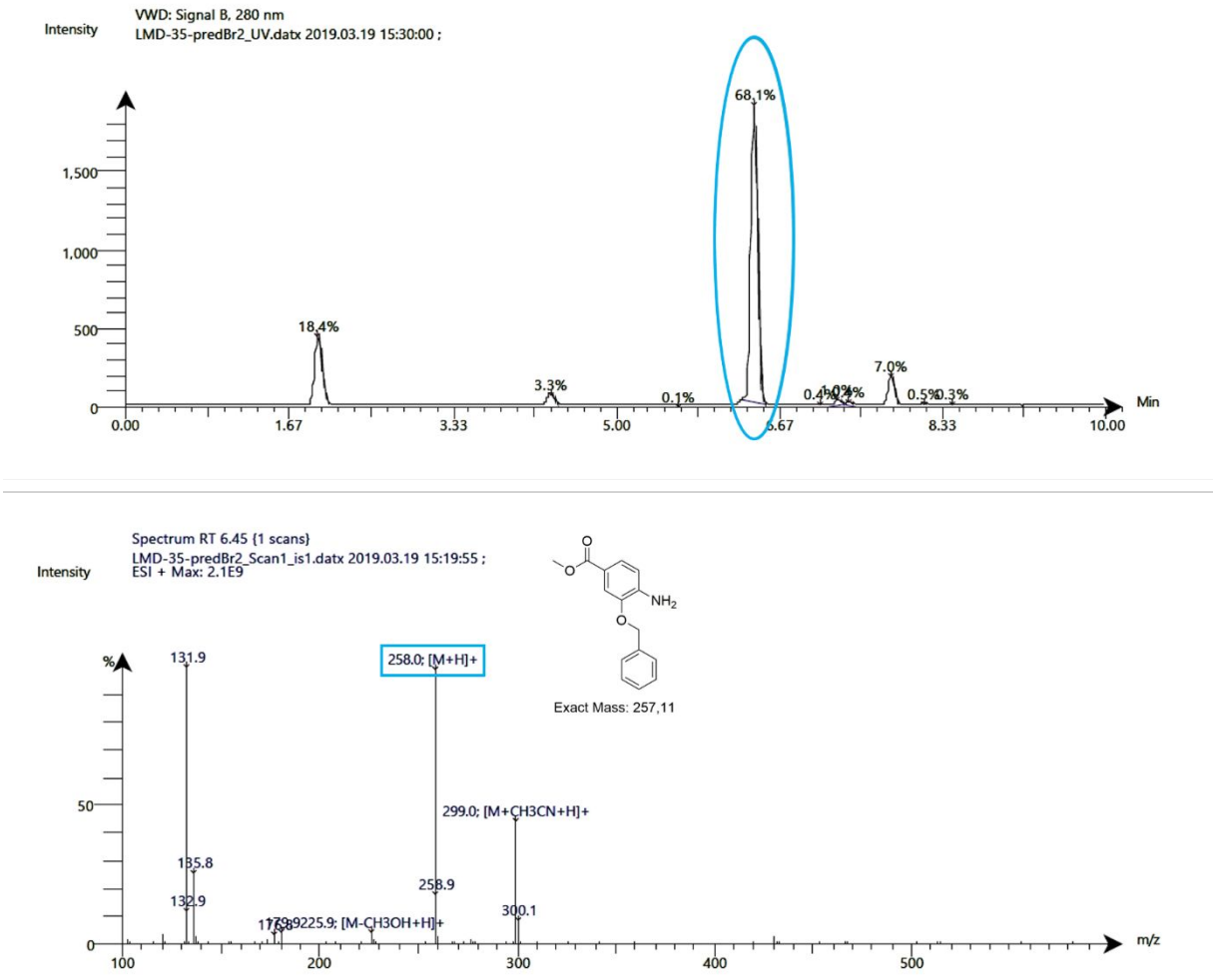
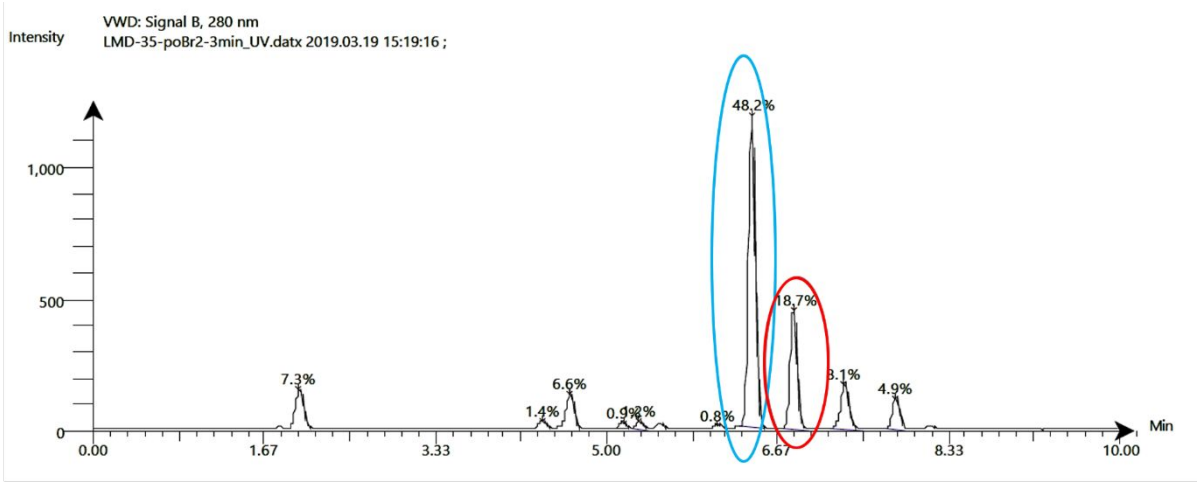
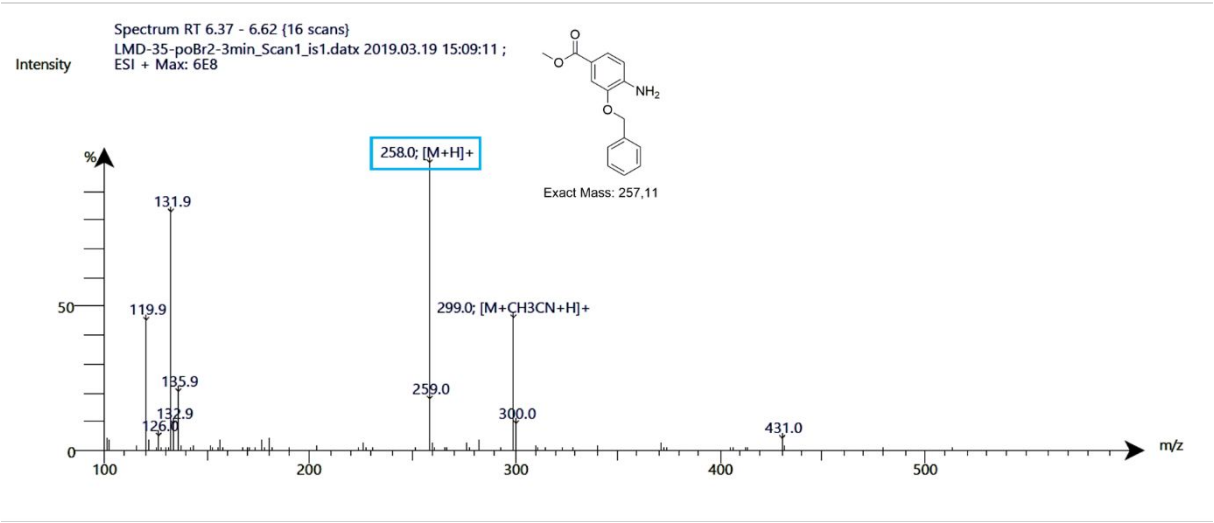


Figure S2. HPLC-MS analysis of the reaction mixture 3 minutes after the addition of bromine (starting compound **4b** and intermediate **I1**).





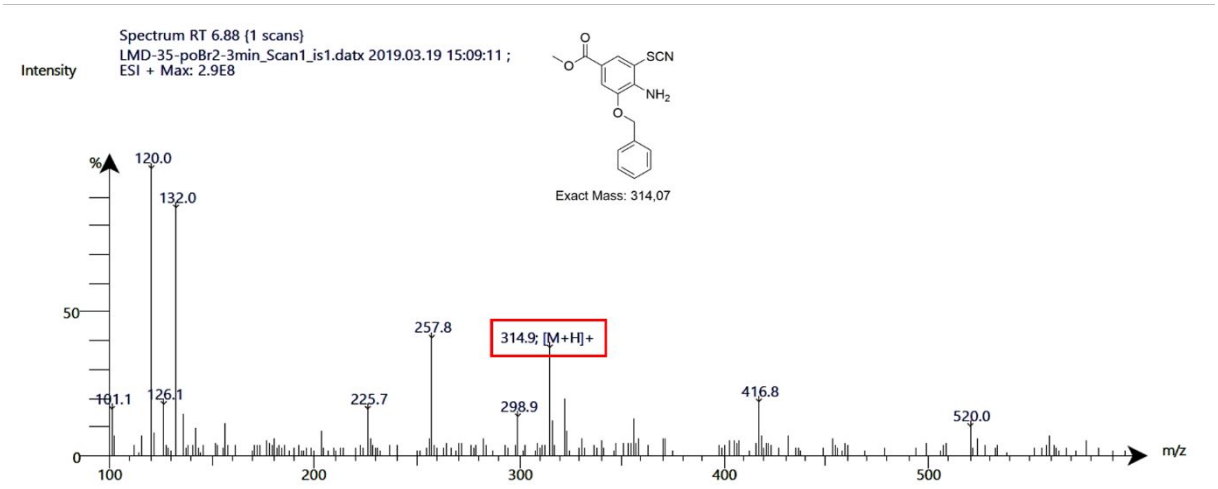


Figure S3. HPLC-MS analysis of the reaction mixture 30 minutes after the addition of bromine (intermediate **I1**).

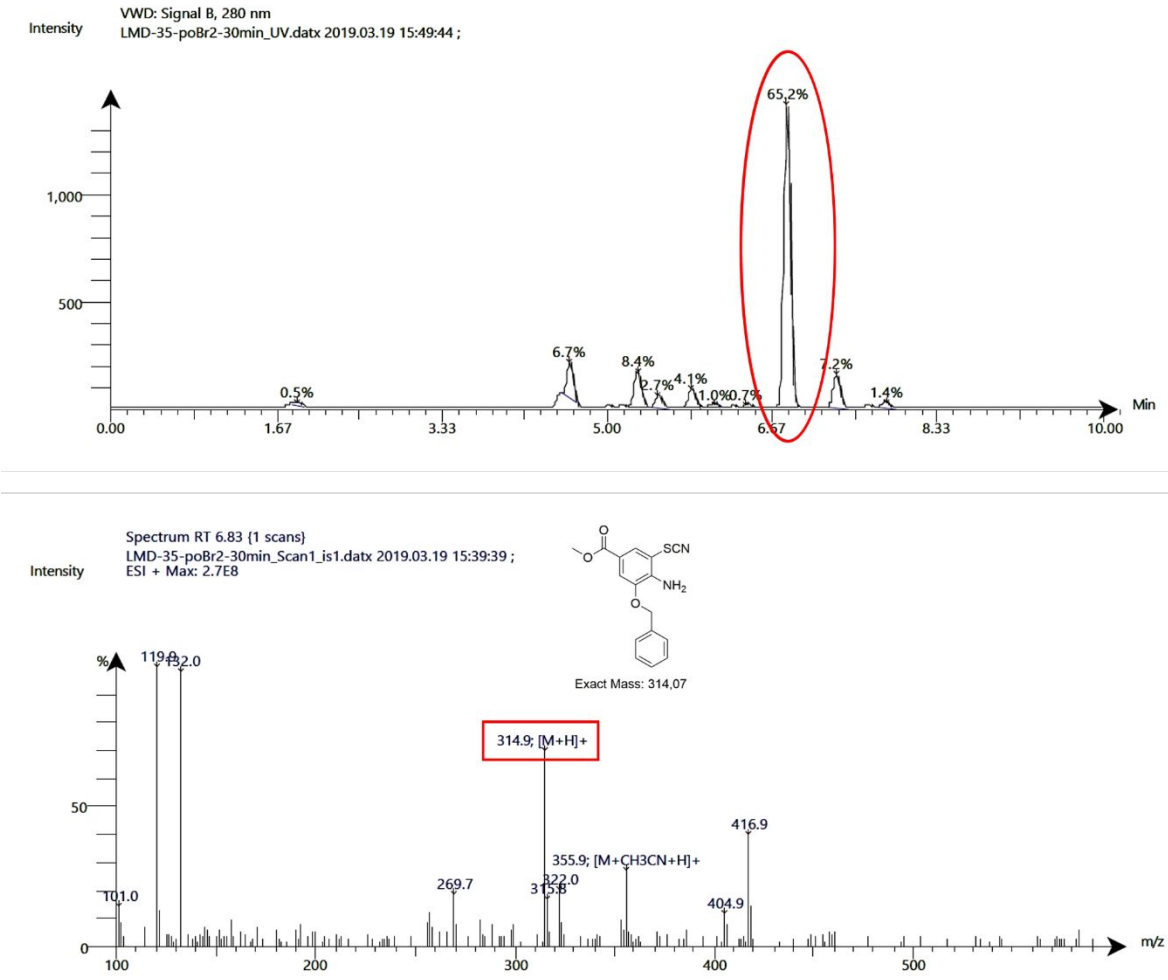


Figure S4. NMR spectra of intermediate **I1** (reaction mixture 30 minutes after the addition of bromine).

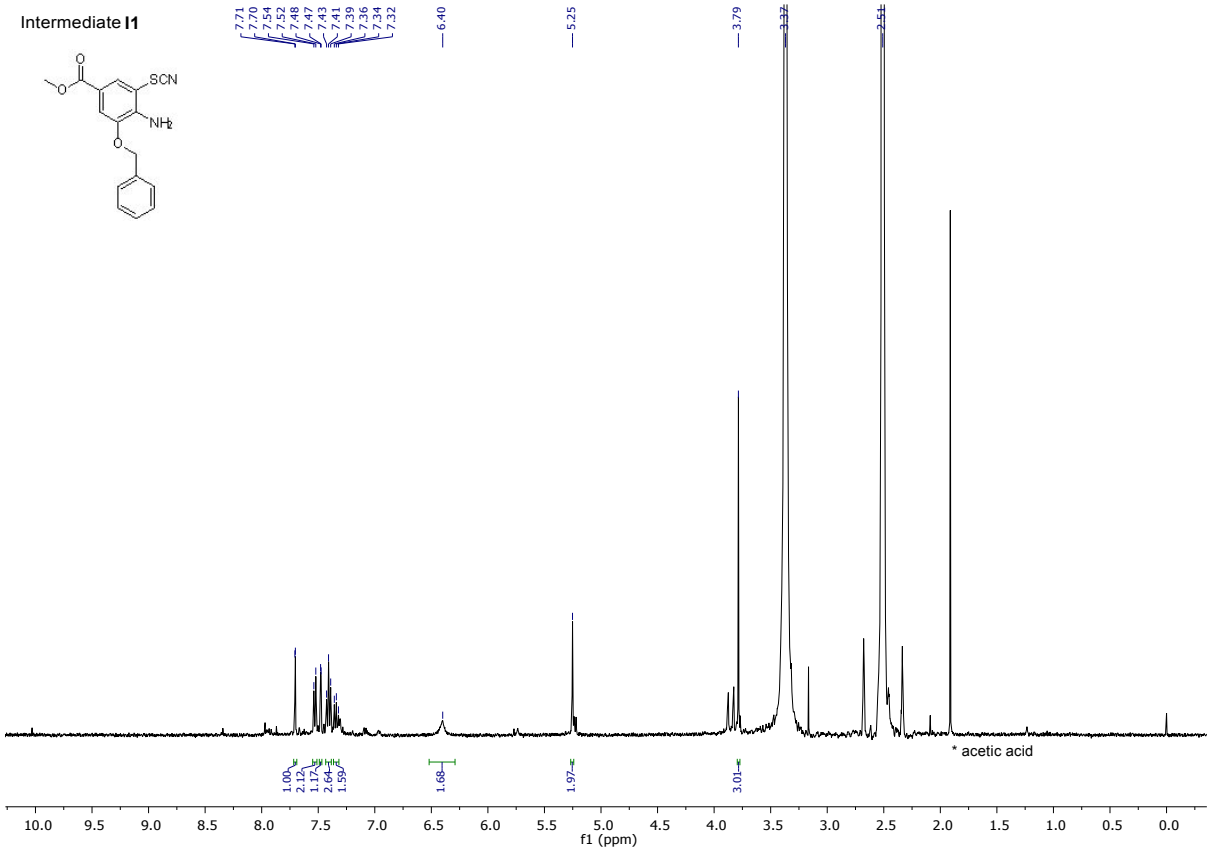


Figure S5. HPLC-MS analysis of the reaction mixture 60 minutes after the addition of bromine (intermediate **I1** and product **5b**).

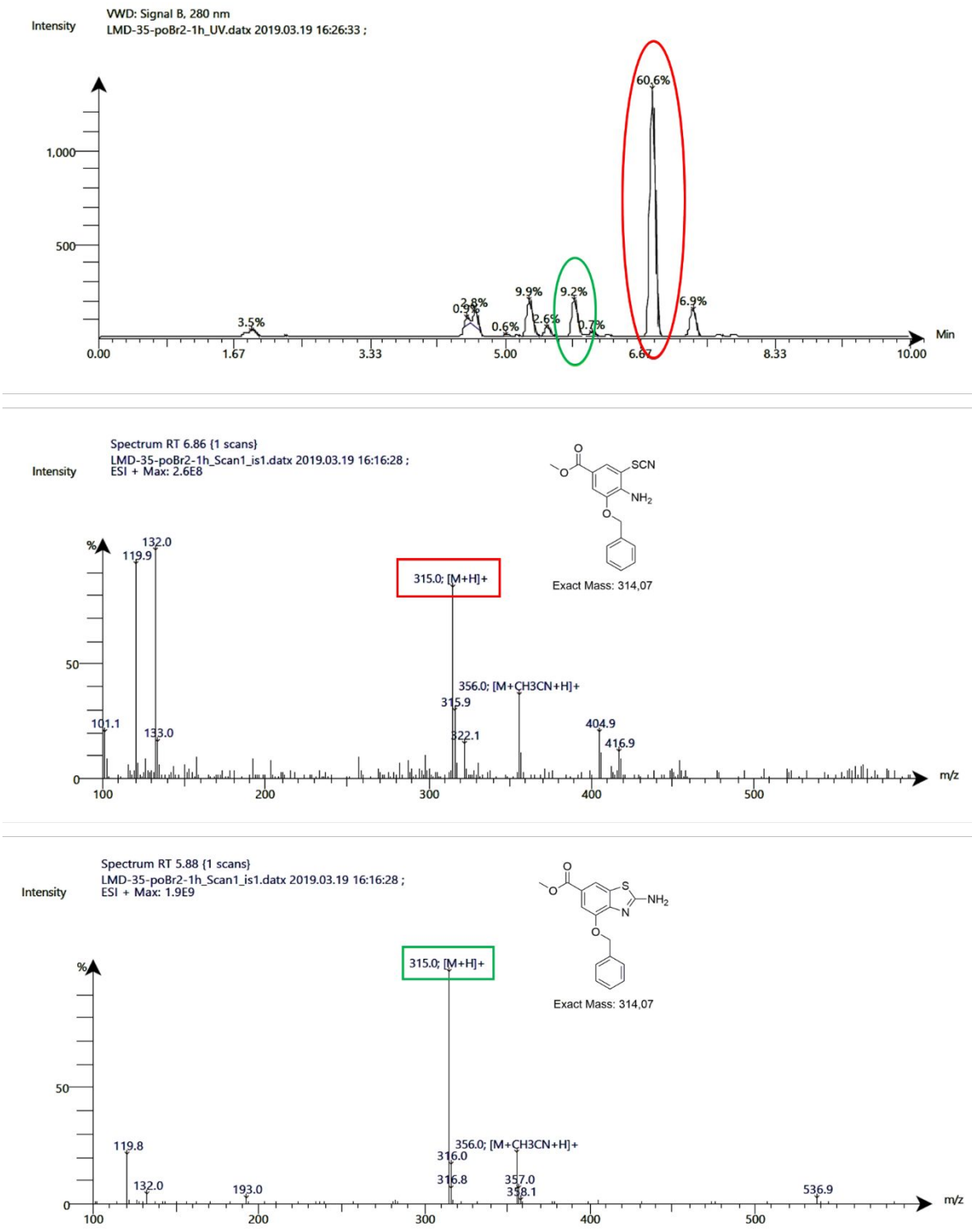


Figure S6. Stacked NMR spectra of compound **4b**, intermediate **I1** and compound **5b**.

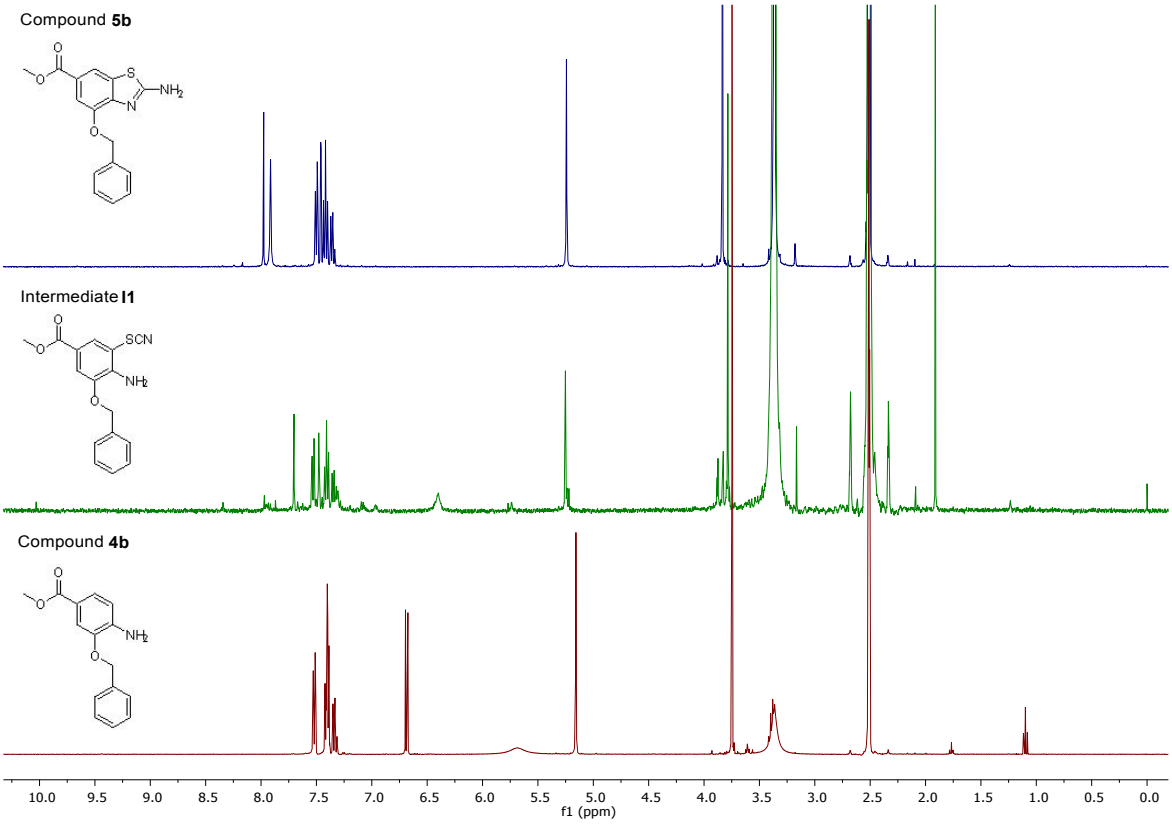
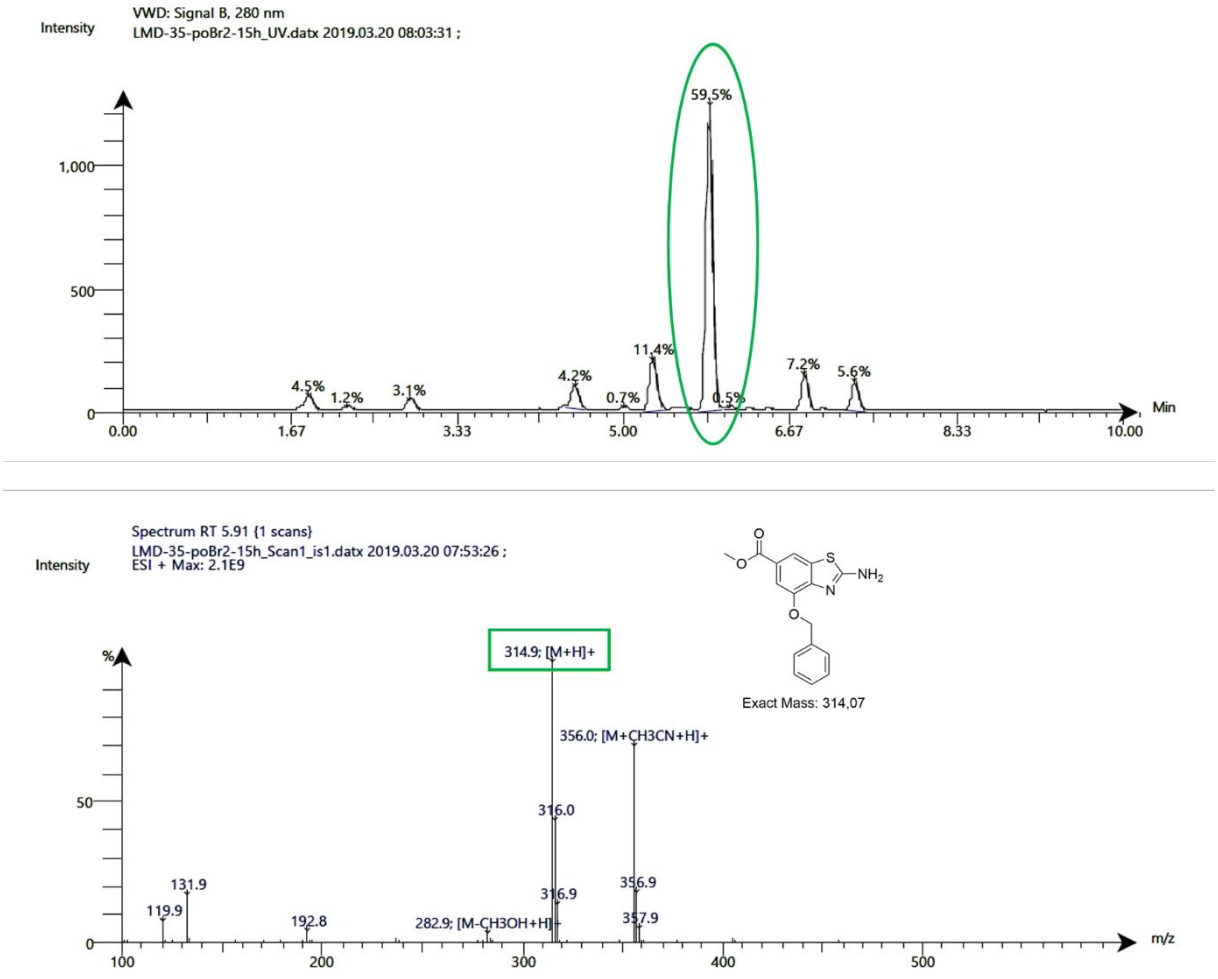


Figure S7. HPLC-MS analysis of the reaction mixture 15 hours after the addition of bromine (product **5b**).

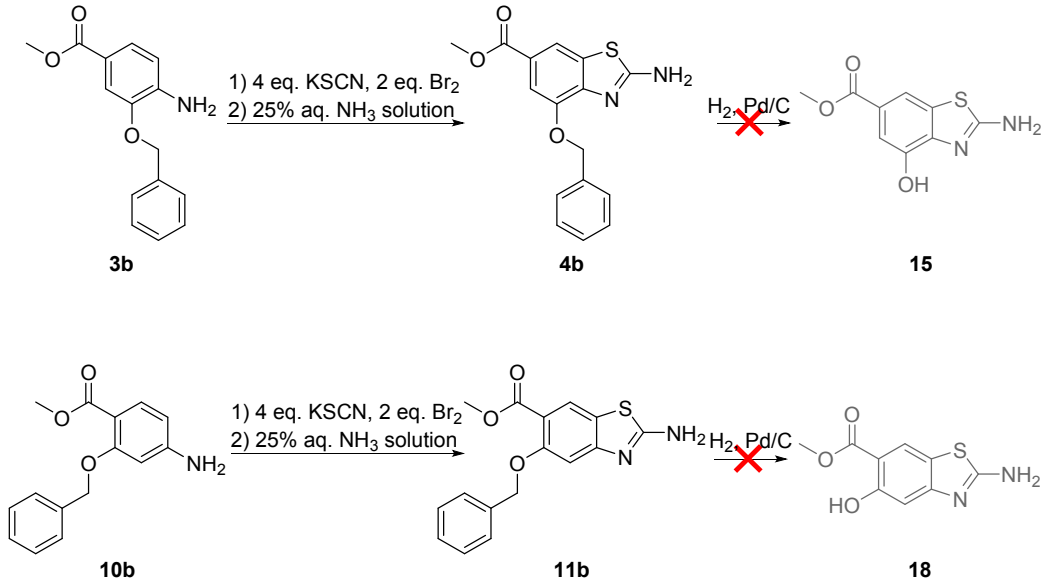


S2. Optimization of synthesis of methyl 2-amino-4- or -5-hydroxybenzo[*d*]thiazole-6-carboxylates

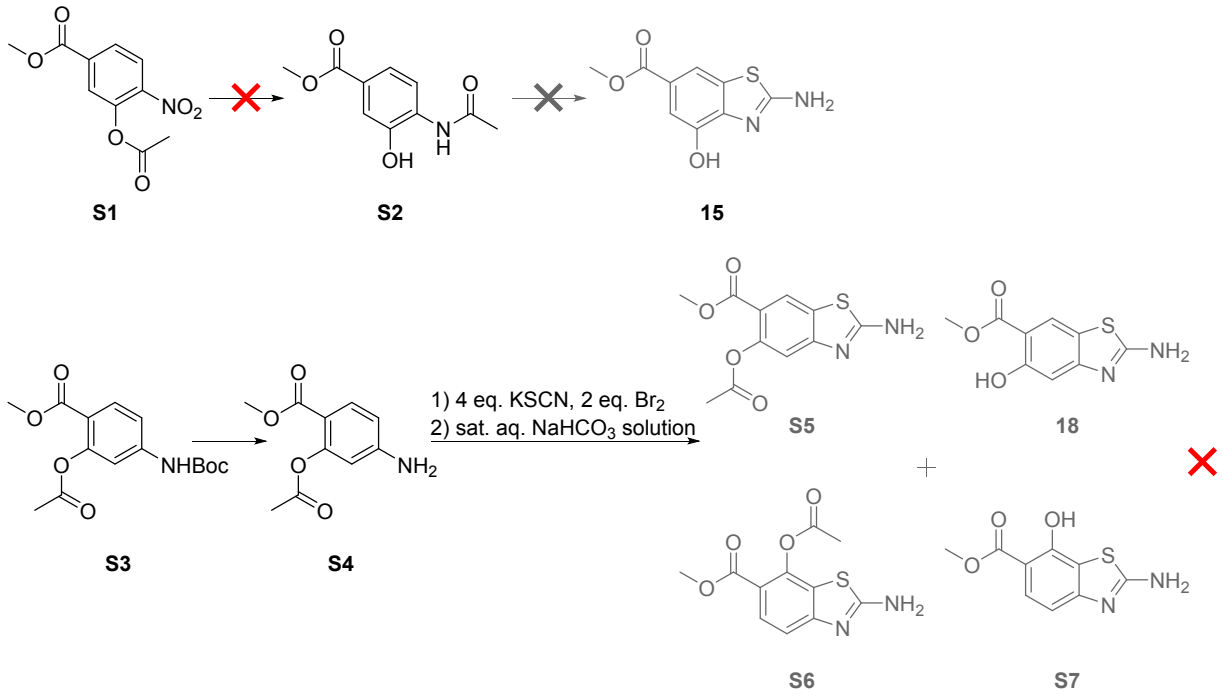
S2.1 Exploration of benzyl and acetyl protecting groups

The first approach for the protection of OH group was benzyl protecting group (i.e., **5b**, **11b**). Although it offered a suitable protection in the cyclisation step, its removal with catalytic hydrogenation was not successful (SI, Scheme S1). The next approach was the use of an acetyl protecting group to protect the methyl 3-hydroxy-4-nitrobenzoate (**S1**; SI, Scheme S2), which was again not successful, as the acetyl group migrated from OH to NH_2 after reduction of the nitro group (**S2**; SI, Scheme S2). In the case of the Boc-protected methyl 2-acetoxy-4-aminobenzoate (**S3**; SI, Scheme S2), the acetyl group did not move after the removal of the Boc protecting group (**S4**; SI, Scheme S2). However, the cyclization reaction in the next step was not regioselective, and thus a mixture of compounds with hydroxyl groups at positions 5 and 7 of the benzothiazole was formed (**S5-7**, **18**; SI, Scheme S2) in which some of the compounds were partly still acetyl protected, while with some of them the acetyl was already removed during isolation (SI, Scheme S2).

Scheme S1. Optimization of synthesis of methyl 2-amino-4- or -5-hydroxybenzo[*d*]thiazole-6-carboxylates: benzyl protecting group.



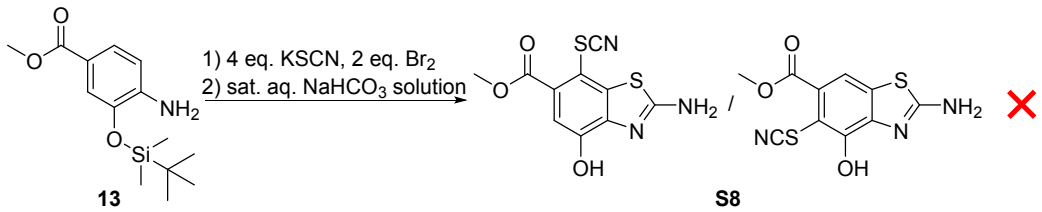
Scheme S2. Optimization of synthesis of methyl 2-amino-4- or -5-hydroxybenzo[*d*]thiazole-6-carboxylates: acetyl protecting group.



S2.2 Exploration of *tert*-butyldimethylsilyl protecting group: OH group at position 4 of the benzo[*d*]thiazole bicycle

In the third attempt the *tert*-butyldimethylsilyl group was used for protection of the hydroxyl group. A cyclization reaction with methyl 4-amino-3-((*tert*-butyldimethylsilyl)oxy)benzoate was set up following the general procedure but instead of ammonia in the work-up phase, saturated aqueous $NaHCO_3$ was used for the neutralization. As a result, a cyclized product with an unsubstituted OH group was obtained (**S8**; SI, Scheme S3), thus completing both the cyclization and the removal of the protecting group in one reaction step. However, the product was additionally substituted with the SCN group on the benzene ring, and so it could not be used in the following reactions (SI, Scheme S3). As the presence of a silyl ether group apparently highly activates the system for electrophilic aromatic substitution, to potentially avoid the additional substitution, the amounts of KSCN and bromine were halved (from initial 4 equiv. of KSCN and 2 equiv. of Br_2). Additionally, in a second approach, ammonia solution was again used for the neutralization (Scheme 5). In this way, we successfully prepared a *tert*-butyldimethylsilyl-protected product **14** (Scheme 5). Finally, we combined the two approaches – reduced amount of KSCN and bromine, and saturated aq. $NaHCO_3$ as the base – and thus successfully obtained the desired product with an unsubstituted OH group at position 4 (**15**; Scheme 5).

Scheme S3. Optimization of synthesis of methyl 2-amino-4-hydroxybenzo[*d*]thiazole-6-carboxylate: *tert*-butyldimethylsilyl protecting group.



S2.3 Exploration of *tert*-butyldimethylsilyl protecting group: OH group at position 5 of the benzo[*d*]thiazole bicycle

To optimize the reaction conditions used to prepare 5-substituted products and to selectively synthesize only the *tert*-butyldimethylsilyl protected product **17** or only the deprotected product **18**, the nature of the base used during the work-up was modified. However, the product was partly deprotected when using 25% NH_3 solution, even if the neutralization was performed rapidly and on ice. On the other hand, the use of saturated aq. $NaHCO_3$ did not yield only the

deprotected product, even after a long neutralization process. The use of a stronger base such as 1 M NaOH was avoided because of the presence of the methyl ester. Nevertheless, it was easy to separate the mixture of *O*-protected and *O*-unprotected products obtained after the reaction (see SI, Section S4 for more detailed procedures).

S3. General experimental details

Chemicals were obtained from Acros Organics (Geel, Belgium), Sigma-Aldrich (St. Louis, MO, USA) and Apollo Scientific (Stockport, UK) and used without further purification. Analytical TLC was performed on silica gel Merck 60 F254 plates (0.25 mm), using visualization with UV light and spray reagents. Column chromatography was carried out on silica gel 60 (particle size 240–400 mesh). ^{1}H and ^{13}C NMR spectra were recorded at 400 and 100 MHz, respectively, on a Bruker AVANCE III 400 spectrometer (Bruker Corporation, Billerica, MA, USA) in DMSO-d_6 or $CDCl_3$ solutions, with TMS as the internal standard. HPLC-MS analyses were performed on an Agilent Technologies 1260 Infinity II LC System (Agilent Technologies, Inc., Santa Clara, CA, USA) coupled to an ADVION expression CMSL mass spectrometer (Advion Inc., Ithaca, USA). The column used was Waters XBridge C18 column (3.5 μm, 4.6 mm × 150 mm), a flow rate of 1.5 mL/min and sample injection volume of 10 μL. Mobile phase consisted of acetonitrile (as solvent A) and 0.1% formic acid and 1% acetonitrile in ultrapure water (as solvent B). The gradient (for solvent A) was: 0-1 min, 25%; 1-6 min, 25%-98%%; 6-6.5 min, 98%; 6.5-7.5 min, 98%-25%; 7.5-10.5 min, 25%. Mass spectra were obtained using Exactive Plus Orbitrap mass spectrometer (Thermo Fisher Scientific, Waltham, Massachusetts, ZDA) or ADVION expression CMS^{L} mass spectrometer (Advion Inc., Ithaca, USA).

S4. Synthesis and characterization of compounds

Methyl 3-(benzyloxy)-4-nitrobenzoate (3b).[1] Synthesized according to General procedure C with benzyl bromide (0.788 mL, 6.62 mmol) as a reagent and acetonitrile (20 mL) as solvent. Yield: 1.54 g (90%); yellow crystals. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.92 (s, 3H), 5.41 (s, 2H), 7.29-7.51 (m, 5H), 7.70 (dd, J = 8.3, 1.6 Hz, 1H), 7.90 (d, J = 1.5 Hz, 1H), 8.03 (d, J = 8.3 Hz, 1H).

Methyl 3-(2-morpholinoethoxy)-4-nitrobenzoate (3c). Synthesized according to General procedure C with Na_2CO_3 (538 mg, 5.07 mmol) and 4-(2-chloroethyl)morpholine hydrochloride (566 mg, 3.04 mmol) as reagents. The reaction mixture was stirred at 80 °C. Yield: 760 mg (97%); yellow crystals. ^{1}H NMR (400 MHz, DMSO-d_6) δ 2.47 (s, 4H), 2.72 (t,

J = 5.2 Hz, 2H), 3.50-3.60 (m, 4H), 3.91 (s, 1H), 4.37 (t, *J* = 5.5 Hz, 2H), 7.67 (dd, *J* = 8.3, 1.6 Hz, 1H), 7.85 (d, *J* = 1.6 Hz, 1H), 7.99 (d, *J* = 8.3 Hz, 1H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 53.3, 54.0, 57.1, 66.6, 68.5, 116.3, 121.8, 125.5, 134.7, 143.0, 151.3, 165.2.

Methyl 3-(2-((*tert*-butoxycarbonyl)amino)ethoxy)-4-nitrobenzoate (3d).[2] Synthesized according to General procedure C with *tert*-butyl (2-chloroethyl)carbamate (0.957 g, 5.33 mmol) as a reagent and the addition of KI (1.18 g, 7.10 mmol). The reaction mixture was stirred at 80 °C. The crude product was purified with flash column chromatography using ethyl acetate/hexane (1:3) as eluent. Yield: 260 mg (22%); yellow crystals. ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.37 (s, 9H), 3.28-3.33 (m, 2H), 3.90 (s, 3H), 4.24 (t, *J* = 5.6 Hz, 2H), 6.96 (t, *J* = 5.3 Hz, 1H), 7.66 (dd, *J* = 8.3, 1.6 Hz, 1H), 7.78 (d, *J* = 1.4 Hz, 1H), 7.99 (d, *J* = 8.3 Hz, 1H).

Methyl 3-((3-(methoxycarbonyl)benzyl)oxy)-4-nitrobenzoate (3e). Synthesized according to General procedure C with methyl 3-(bromomethyl)benzoate (559 mg, 2.44 mmol) as a reagent and acetonitrile (15 mL) as solvent. Yield: 535 g (76%); white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.87 (s, 3H), 3.91 (s, 3H), 5.50 (s, 2H), 7.59 (t, *J* = 7.7 Hz, 1H), 7.71 (dd, *J* = 8.4, 1.6 Hz, 1H), 7.74 (dt, *J* = 7.8, 1.5 Hz, 1H), 7.92 (d, *J* = 1.6 Hz, 1H), 7.95 (dt, *J* = 7.7, 1.5 Hz, 1H), 8.05 (d, *J* = 8.4 Hz, 1H), 8.10 (t, *J* = 1.7 Hz, 1H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 52.7, 53.4, 70.6, 116.3, 122.2, 125.8, 128.6, 129.4, 129.6, 130.4, 132.6, 134.8, 137.0, 142.9, 150.9, 165.2, 166.4.

Methyl 4-amino-3-(2-morpholinoethoxy)benzoate (4c). Synthesized according to General procedure D using methyl 3-(2-morpholinoethoxy)-4-nitrobenzoate (**3c**, 750 mg, 2.42 mmol). Yield: 672 mg (99%); off-green crystals. ^{1}H NMR (400 MHz, DMSO-d_6) δ 2.46-2.50 (m, 4H), 2.64-2.80 (m, 2H), 3.54-3.66 (m, 4H), 3.75 (s, 3H), 4.08 (t, *J* = 5.5 Hz, 2H), 5.68 (s, 2H), 6.65 (d, *J* = 8.2 Hz, 1H), 7.34 (d, *J* = 1.8 Hz, 1H), 7.40 (dd, *J* = 8.2, 1.8 Hz, 1H).

Methyl 4-amino-3-(2-((*tert*-butoxycarbonyl)amino)ethoxy)benzoate (4d).[3] Synthesized according to General procedure D using methyl 3-(2-((*tert*-butoxycarbonyl)amino)ethoxy)-4-nitrobenzoate (250 mg, 0.735 mmol). Yield: 220 mg (97%); green oily product. ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.40 (s, 9H), 3.36 (m, 2H, signal is overlapping with the signal for water), 3.75 (s, 3H), 3.90 (t, *J* = 5.1 Hz, 2H), 5.80 (s, 2H), 6.62 (d, *J* = 8.2 Hz, 1H), 7.18 (t, *J* = 6.0 Hz, 1H), 7.23 (d, *J* = 1.7 Hz, 1H), 7.38 (dd, *J* = 8.2, 1.8 Hz, 1H).

Methyl 4-amino-3-((3-(methoxycarbonyl)benzyl)oxy)benzoate (4e). Synthesized according to General procedure E using methyl 3-((3-(methoxycarbonyl)benzyl)oxy)-4-nitrobenzoate (333 mg, 0.964 mmol). Yield: 200 mg (66%); pale yellow crystals. ^{1}H NMR (400 MHz, DMSO-

d_6) δ 3.75 (s, 3H), 3.87 (s, 3H), 5.24 (s, 2H), 5.71 (s, 2H), 6.69 (d, *J* = 8.7 Hz, 1H), 7.40-7.42 (m, 2H), 7.57 (t, *J* = 7.7 Hz, 1H), 7.81-7.86 (m, 1H), 7.91-7.95 (m, 1H), 8.08-8.09 (m, 1H).

Methyl 4-amino-3-(2-methoxyethoxy)benzoate (4f).[4] Synthesized according to General procedure D using methyl 3-(2-methoxyethoxy)-4-nitrobenzoate (1.00 g, 3.92 mmol). The crude product was purified with flash column chromatography using hexane/ethyl acetate (1:1) as eluent. Yield: 640 mg (80%, over two steps); transparent oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.44 (s, 3H), 3.74-3.80 (m, 2H), 3.85 (s, 3H), 4.16-4.23 (m, 2H), 4.31 (s, 2H), 6.67 (d, *J* = 8.2 Hz, 1H), 7.47 (d, *J* = 1.7 Hz, 1H), 7.56 (dd, *J* = 8.2, 1.8 Hz, 1H).

Methyl 2-amino-4-methoxybenzo[*d*]thiazole-6-carboxylate (5a).[5] Synthesized according to General procedure A using methyl 4-amino-3-methoxybenzoate (2.34 g, 12.9 mmol). The product was obtained after filtration of the precipitate out of ethyl acetate/water mixture. Yield: 2.72 g (88%); orange solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.84 (s, 3H), 3.88 (s, 3H), 7.35 (d, *J* = 1.5 Hz, 1H), 7.85 (s, 2H), 7.96 (d, *J* = 1.6 Hz, 1H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 51.9, 55.7, 108.2, 115.6, 122.2, 131.6, 146.3, 149.0, 166.1, 168.6. HRMS (ESI$^+$) m/z for $C_{10}H_{11}N_2O_3S$ ([M+H]$^+$): calculated 239.0485, found 239.0488.

Methyl 2-amino-4-(benzyloxy)benzo[*d*]thiazole-6-carboxylate (5b). Synthesized according to General procedure A using methyl 4-amino-3-(benzyloxy)benzoate (229 mg, 0.89 mmol). After the solid was filtered and washed with water it was dried and dispersed in methanol. The suspension was sonicated, heated and filtered hot. The process was repeated three times and the solvent of the mother liquid was removed *in vacuo* to afford the product as yellow solid. Yield: 176 mg (63%); yellow solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.83 (s, 3H), 5.24 (s, 2H), 7.32-7.44 (m, 3H), 7.46 (d, *J* = 1.6 Hz, 1H), 7.48-7.53 (m, 2H), 7.91 (s, 2H), 7.98 (d, *J* = 1.5 Hz, 1H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 51.9, 70.0, 109.9, 115.9, 122.2, 127.9, 128.3, 131.8, 137.0, 146.6, 148.0, 166.1, 168.7. HRMS (ESI$^+$) m/z for $C_{16}H_{15}N_2O_3S$ ([M+H]$^+$): calculated 315.0798, found 315.0804.

Methyl 2-amino-4-(2-morpholinoethoxy)benzo[*d*]thiazole-6-carboxylate (5c). Synthesized according to General procedure A using methyl 4-amino-3-(2-morpholinoethoxy)benzoate (627 mg, 2.24 mmol). Yield: 431 mg (57%); orange solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ 2.55 (s, 4H), 2.80 (s, 2H), 3.61 (s, 4H), 3.83 (s, 3H), 4.25 (s, 2H), 7.39 (s 1H), 7.91 (s, 2H), 7.97 (s, 1H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 51.9, 53.2, 56.7, 65.5, 65.6, 110.3, 116.1, 122.2, 131.9, 146.7, 147.9, 166.1, 168.7. HRMS (ESI$^+$) m/z for $C_{15}H_{20}N_3O_4S$ ([M+H]$^+$): calculated 338.1169, found 338.1178.

Methyl 2-amino-4-(2-((*tert*-butoxycarbonyl)amino)ethoxy)benzo[*d*]thiazole-6-carboxylate (5d). Synthesized according to General procedure A using methyl 4-amino-3-(2-((*tert*-butoxycarbonyl)amino)ethoxy)benzoate (214 mg, 0.690 mmol). After the solid was filtered and washed with water it was dried and dispersed in methanol. The suspension was sonicated, heated and filtered hot. The process was repeated three times and the solvent of the mother liquid was removed *in vacuo* to afford the product as orange solid. Yield: 241 mg (95%); orange solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.39 (s, 9H), 3.33 (q, J = 6.0 Hz, 2H), 3.83 (s, 3H), 4.11 (t, J = 6.0 Hz, 2H), 7.00 (t, J = 5.2 Hz, 1H), 7.36 (d, J = 1.6 Hz, 1H), 7.92 (br s, 2H), 7.97 (d, J = 1.5 Hz, 1H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 28.2, 51.9, 67.2, 77.8, 110.1, 115.9, 122.2, 131.9, 146.6, 148.0, 155.6, 166.1, 168.6, 172.0. HRMS (ESI$^+$) m/z for $C_{16}H_{22}N_3O_5S$ ([M+H]$^+$): calculated 368.1275, found 368.1286.

Methyl 2-amino-4-((3-(methoxycarbonyl)benzyl)oxy)benzo[*d*]thiazole-6-carboxylate (5e). Synthesized according to General procedure A using methyl 4-amino-3-((3-(methoxycarbonyl)benzyl)oxy)benzoate (197 mg, 0.625 mmol). The solid after filtration was purified with flash column chromatography using ethyl acetate/hexane (1:1) as eluent. Yield: 81 mg (35%); white crystals. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.83 (s, 3H), 3.88 (s, 3H), 5.33 (s, 2H), 7.47 (d, J = 1.5 Hz, 1H), 7.58 (m, 1H), 7.79 (m, 1H), 7.94 (s, 2H), 7.95 (m, 1H), 7.99 (d, J = 1.5 Hz, 1H), 8.10 (m, 1H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 52.0, 52.2, 69.4, 110.0, 116.1, 122.2, 128.3, 128.6, 128.9, 129.7, 131.9, 132.6, 137.8, 146.6, 147.8, 166.1, 166.1, 168.8. HRMS (ESI$^+$) m/z for $C_{18}H_{17}N_2O_5S$ ([M+H]$^+$): calculated 373.0853, found 373.0864.

Methyl 2-amino-4-(2-methoxyethoxy)benzo[*d*]thiazole-6-carboxylate (5f). Synthesized according to General procedure A using methyl 4-amino-3-(2-methoxyethoxy)benzoate (300 mg, 1.33 mmol). Yield: 150 mg (40%); yellow solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.34 (s, 3H), 3.66-3.71 (m, 2H), 3.83 (s, 3H), 4.19-4.25 (m, 2H), 7.35 (d, J = 1.6 Hz, 1H), 7.91 (s, 2H), 7.96 (d, J = 1.6 Hz, 1H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 52.4, 58.6, 68.2, 70.9, 109.9, 116.3, 122.7, 132.3, 147.0, 148.6, 166.6, 169.1. HRMS (ESI$^+$) m/z for $C_{12}H_{15}N_2O_4S$ ([M+H]$^+$): calculated 283.0747, found 283.0744.

Methyl 4-amino-2-hydroxybenzoate (7).[6] Synthesized according to General procedure B using 4-aminosalicylic acid (**6**, 7.50 g, 49.0 mmol). Yield: 7.26 g (89%); brown solid; mp: 99-104 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.90 (s, 3H), 4.12 (s, 2H), 6.12-6.20 (m, 2H), 7.64 (dd, J = 8.0, 0.9 Hz, 1H), 10.97 (s, 1H).

Methyl 4-((*tert*-butoxycarbonyl)amino)-2-methoxybenzoate (9a).[7] Synthesized according to General procedure C using methyl 4-((*tert*-butoxycarbonyl)amino)-2-hydroxybenzoate (**8**) as the starting compound. Reaction mixture was stirred at room temperature instead of 60 °C. The crude product was purified with flash column chromatography using ethyl acetate/hexane (1:3) as an eluent. Yield: 0.867 g (92%); white crystals. ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.53 (s, 9H), 3.86 (s, 3H), 3.92 (s, 3H), 6.66 (s, 1H), 6.71 (dd, J = 8.5, 2.1 Hz, 1H), 7.39 (d, J = 2.0 Hz, 1H), 7.79 (d, J = 8.5 Hz, 1H).

Methyl 2-(benzyloxy)-4-((*tert*-butoxycarbonyl)amino)benzoate (9b). Synthesized according to General procedure C using methyl 4-((*tert*-butoxycarbonyl)amino)-2-hydroxybenzoate (**8**) as the starting compound and benzyl bromide (0.667 mL, 5.61 mmol) as reagent and stirring the reaction mixture at room temperature instead of 60 °C. To the crude product hexane was added, the obtained suspension sonicated, the solid was filtered off, washed with hexane and dried. Yield: 0.725 g (54%); white solid. ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.53 (s, 9H), 3.87 (s, 3H), 5.19 (s, 2H), 6.65 (s, 1H), 6.75 (dd, J = 8.5, 2.0 Hz, 1H), 7.29-7.32 (m, 1H), 7.37-7.41 (m, 2H), 7.44 (s, 1H), 7.53-7.55 (m, 2H), 7.83 (d, J = 8.5 Hz, 1H).

Methyl 2-amino-5-methoxybenzo[*d*]thiazole-6-carboxylate (11a). Synthesized according to General procedure A using methyl 4-amino-2-methoxybenzoate **(10a**, 0.615 g, 3.39 mmol) as a reagent. The precipitate obtained after filtration out of ammonium water mixture was suspended in methanol, heated and the undissolved residue filtered off. The mother liquid was evaporated under reduced pressure and the crude product suspended in diethyl ether (40 mL), sonicated, filtered and dried. Yield: 0.600 g (74%); yellow solid. mp: 145-149 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.70 (s, 3H), 3.77 (s, 3H), 6.46 (s, 1H), 6.70 (s, 2H), 7.86 (s, 1H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 51.8, 56.1, 94.3, 97.8, 108.5, 112.2, 141.6, 155.6, 163.3, 164.7. MS (ESI) m/z = 261.0 ($[M+Na]^+$).

Methyl 2-amino-5-(benzyloxy)benzo[*d*]thiazole-6-carboxylate (11b). Synthesized according to General procedure A using methyl 4-amino-2-(benzyloxy)benzoate **(10b**, 210 mg, 0.817 mmol) as a reagent. The precipitate obtained after filtration out of ammonium water mixture was suspended in methanol, heated and the undissolved residue filtered off. The mother liquid was evaporated under reduced pressure and the crude product suspended in diethyl ether (40 mL), sonicated, filtered and dried. Yield: 115 mg (45%); yellow solid. mp: 133-136 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.74 (s, 3H), 5.15 (s, 2H), 6.56 (s, 1H), 6.72 (s, 2H), 7.28-7.38 (m, 1H), 7.40-7.44 (m, 2H), 7.50-7.53 (m, 2H), 7.92 (s, 1H). ^{13}C NMR (101 MHz, DMSO-d_6)

δ 51.8, 69.8, 94.7, 99.0, 108.7, 112.2, 127.3, 128.1, 128.8, 137.0, 141.7, 155.6, 162.3, 164.6. MS (ESI) m/z = 337.1 ([M+Na]$^+$).

Methyl 4-amino-3-((*tert*-butyldimethylsilyl)oxy)benzoate (13).[8] Synthesized according to General procedure D using crude methyl 3-((*tert*-butyldimethylsilyl)oxy)-4-nitrobenzoate (**12**, 2.8 g). Crude product was purified using flash column chromatography using ethyl acetate/hexane (1:2) as eluent. Yield: 1.62 g (56%, over two steps); white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ 0.22 (s, 6H), 0.98 (s, 9H), 3.74 (s, 3H), 5.39 (s, 2H), 6.70 (d, J = 8.3 Hz, 1H), 7.23 (d, J = 1.9 Hz, 1H), 7.38 (dd, J = 8.3, 1.9 Hz, 1H).

Methyl 2-amino-4-((*tert*-butyldimethylsilyl)oxy)benzo[*d*]thiazole-6-carboxylate (14). Synthesized according to General procedure A using methyl 4-amino-3-((*tert*-butyldimethylsilyl)oxy)benzoate (1.0 g, 3.55 mmol). Yield: 726 mg (60%); yellow solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ 0.22 (s, 6H), 0.99 (s, 9H), 3.81 (s, 3H), 7.26 (d, J = 1.6 Hz, 1H), 7.79 (s, 2H), 7.92 (d, J = 1.7 Hz, 1H). ^{13}C NMR (101 MHz, DMSO-d_6) δ -4.2, 18.3, 25.7, 52.0, 116.1, 117.0, 122.3, 132.5, 144.6, 149.2, 166.0, 168.1. HRMS (ESI$^+$) m/z for $C_{15}H_{23}N_2O_3SSi$ ([M+H]$^+$): calculated 339.1193, found 339.1191.

Methyl 2-amino-4-hydroxybenzo[*d*]thiazole-6-carboxylate (15). Synthesized according to General procedure A using methyl 4-amino-3-((*tert*-butyldimethylsilyl)oxy)benzoate (**7**, 1.00 g, 3.55 mmol) and only 2 equivalents of KSCN (0.70 g, 7.11 mmol) and 1 equivalent of bromine (0.183 mL, 3.55 mmol). To isolate the product, the reaction mixture was neutralized with saturated aqueous $NaHCO_3$ solution (350 mL), the obtained suspension was extracted with ethyl acetate (3 × 100 mL), and the combined organic phases washed again with saturated aq. $NaHCO_3$ solution (70 mL) and brine (70 mL). The organic phase was dried over Na_2SO_4, filtered and the solvent removed in vacuo. Yield: 230 mg (29%); yellow solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.81 (s, 3H), 7.28 (d, J = 1.6 Hz, 1H), 7.69 (s, 2H), 7.78 (d, J = 1.6 Hz, 1H), 9.66 (s, 1H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 52.3, 112.7, 224.2, 123.0, 132.4, 146.1, 147.5, 166.7, 168.1. HRMS (ESI$^+$) m/z for $C_9H_9N_2O_3S$ ([M+H]$^+$): calculated 225.0328, found 225.0333.

Methyl 2-amino-5-((*tert*-butyldimethylsilyl)oxy)benzo[*d*]thiazole-6-carboxylate (17) and methyl 2-amino-5-hydroxybenzo[*d*]thiazole-6-carboxylate (18).

Synthesized according to General procedure A using methyl 4-amino-2-((*tert*-butyldimethylsilyl)oxy)benzoate (**16**, 4.00 g, 14.2 mmol) and only 2 equivalents of KSCN (2.76 g, 28.4 mmol) and 1 equivalent of bromine (732 µL, 14.2 mmol). To isolate the product, the

reaction mixture was neutralized to pH 8 with saturated aqueous $NaHCO_3$ solution (500 mL) and when EtOAc (150 mL) was added precipitate was formed. The obtained precipitate was filtered off, washed with EtOAc (10 mL) and dried to give unprotected product methyl 2-amino-5-hydroxybenzo[*d*]thiazole-6-carboxylate (**18**) (421 mg) as off-white solid.

The phases of mother liquid from filtration were separated, organic phase was evaporated *in vacuo* and the residue was purified with flash column chromatography using ethyl acetate/hexane (1:4) as eluent to give compound **17** (550 mg) as off-white solid.

Methyl 2-amino-5-((*tert*-butyldimethylsilyl)oxy)benzo[*d*]thiazole-6-carboxylate (17). Mass: 550 mg; off-white solid. mp: 119-123 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 0.19 (s, 6H), 0.99 (s, 9H), 3.76 (s, 3H, CH_3), 6.79 (s, 1H), 7.85 (s, 2H), 8.01 (s, 1H). ^{13}C NMR (101 MHz, DMSO-d_6) δ -4.1, 18.5, 26.1, 51.9, 109.7, 124.0, 166.4, 170.9. MS (ESI) m/z = 339.2 ($[M+H]^+$). HRMS (ESI^+) m/z for $C_{15}H_{23}N_2O_3SSi$ ($[M+H]^+$): calculated 339.1193, found 339.1189.

Methyl 2-amino-5-hydroxybenzo[*d*]thiazole-6-carboxylate (18). Yield 421 mg (13%); off-white solid. ^{1}H NMR (400 MHz, DMSO-d_6): δ 3.88 (s, 3H), 6.82 (s, 1H), 8.01 (s, 2H), 8.08 (s, 1H), 10.67 (s, 1H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 52.7, 104.9, 106.3, 122.6, 123.1, 160.1, 160.2, 170.1, 171.8. HRMS (ESI^+) m/z for $C_9H_9N_2O_3S$ ($[M+H]^+$): calculated 225.0328, found 225.0329.

Methyl 2-amino-4-(benzyloxy)benzo[*d*]thiazole-6-carboxylate (5b). Synthesized according to General procedure C using methyl 2-amino-4-hydroxybenzo[*d*]thiazole-6-carboxylate (**9**, 675 mg, 3.01 mmol), benzyl bromide (375 µL, 3.16 mmol) and K_2CO_3 (832 mg, 6.02 mmol) as reagents. The crude product after extraction was recrystallized from ethyl acetate. Yield: 230 mg (35%); yellow crystals. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.83 (s, 3H), 5.24 (s, 2H), 7.32-7.44 (m, 3H), 7.45 (d, J = 1.6 Hz, 1H), 7.47-7.53 (m, 2H), 7.91 (s, 2H), 7.97 (d, J = 1.5 Hz, 1H).

Methyl 2-amino-4-((4-fluorobenzyl)oxy)benzo[*d*]thiazole-6-carboxylate (19a). Synthesized according to General procedure C using methyl 2-amino-4-hydroxybenzo[*d*]thiazole-6-carboxylate (**15**, 129 mg, 0.575 mmol), 1-(bromomethyl)-4-fluorobenzene (75.3 µL, 0.604 mmol) and K_2CO_3 (159 mg, 1.15 mmol) as reagents. After extraction the product precipitated out of ethyl acetate, the precipitate was filtered off and dried. Yield: 59 mg (31%); pale yellow solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.83 (s, 3H), 5.22 (s, 2H), 7.21-7.29 (m, 2H), 7.45 (d, J = 1.5 Hz, 1H), 7.51-7.58 (m, 2H), 7.92 (s, 2H), 7.98 (d, J = 1.5 Hz, 1H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 52.4, 69.7, 110.4, 115.5, 115.8, 116.5, 122.6, 130.5, 130.6, 132.3, 133.7, 133.7,

147.2, 148.4, 161.0, 163.5, 166.6, 169.2. HRMS (ESI$^+$) m/z for $C_{16}H_{14}FN_2O_3S$ ([M+H]$^+$): calculated 333.0704, found 333.0712.

Methyl 2-amino-4-((3-fluorobenzyl)oxy)benzo[*d*]thiazole-6-carboxylate (19b). Synthesized according to General procedure C using methyl 2-amino-4-hydroxybenzo[*d*]thiazole-6-carboxylate (**15**, 190 mg, 0.847 mmol), 1-(bromomethyl)-3-fluorobenzene (114 µL, 0.932 mmol) and K_2CO_3 (234 mg, 1.69 mmol) as reagents. The crude product after extraction was suspended in diethyl ether, filtered off and dried. Yield: 126 mg (45%); pale yellow solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.82 (s, 3H), 5.26 (s, 2H), 7.13-7.21 (m, 1H), 7.30-7.37 (m, 2H), 7.42-7.49 (m, 2H), 7.93 (s, 2H), 7.98 (d, *J* = 1.5 Hz, 1H). HRMS (ESI$^+$) m/z for $C_{16}H_{14}FN_2O_3S$ ([M+H]$^+$): calculated 333.0704, found 333.0713.

Methyl 2-amino-4-((4-(methoxycarbonyl)benzyl)oxy)benzo[*d*]thiazole-6-carboxylate (19c). Synthesized according to General procedure C using methyl 2-amino-4-hydroxybenzo[*d*]thiazole-6-carboxylate (**15**, 100 mg, 0.446 mmol), methyl 4-(bromomethyl)benzoate (102 mg, 0.446 mmol) and K_2CO_3 (123 mg, 0.892 mmol) as reagents and acetonitrile as solvent. The crude product was recrystallized from methanol. Yield: 69 mg (42%); yellow crystals. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.83 (s, 3H), 3.87 (s, 3H), 5.36 (s, 2H), 7.46 (d, *J* = 1.5 Hz, 1H), 7.65 (d, *J* = 8.3 Hz, 2H), 7.95 (s, 2H), 8.00 (m, 3H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 52.4, 52.7, 69.9, 110.7, 116.6, 122.6, 128.1, 129.5, 129.8, 132.4, 143.1, 147.2, 148.2, 166.5, 169.3. HRMS (ESI$^+$) m/z for $C_{18}H_{17}N_2O_5S$ ([M+H]$^+$): calculated 373.0853, found 373.0862.

Methyl 2-amino-4-(pyrimidin-2-ylmethoxy)benzo[*d*]thiazole-6-carboxylate (19d). Synthesized according to General procedure C using methyl 2-amino-4-hydroxybenzo[*d*]thiazole-6-carboxylate (**15**, 100 mg, 0.446 mmol), 2-(chloromethyl)pyrimidine hydrochloride (81 mg, 0.491 mmol) and K_2CO_3 (123 mg, 0.892 mmol) as reagents and acetonitrile as solvent. The crude product was purified with flash column chromatography using dichloromethane/methanol (9:1) as eluent. Yield: 53 mg (38%); yellow crystals. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.80 (s, 3H), 5.40 (s, 2H), 7.36 (d, *J* = 1.5 Hz, 1H), 7.48 (t, *J* = 4.9 Hz, 1H), 7.89 (s, 2H), 7.95 (d, *J* = 1.5 Hz, 1H), 8.85 (d, *J* = 4.9 Hz, 2H). HRMS (ESI$^+$) m/z for $C_{14}H_{13}N_4O_3S$ ([M+H]$^+$): calculated 317.0703, found 317.0700.

Methyl 2-amino-5-((4-fluorobenzyl)oxy)benzo[*d*]thiazole-6-carboxylate (20). Synthesized according to General procedure C using methyl 2-amino-5-hydroxybenzo[*d*]thiazole-6-carboxylate (**18**, 60 mg, 0.268 mmol), 4-fluorobenzyl bromide (33 µL, 0.268 mmol) and K_2CO_3

(74 mg, 0.535 mmol) as reagents and the reaction mixture was stirred at 80 °C. The crude product was purified with flash column chromatography using dichloromethane/methanol (40:1) as eluent. Yield: 25 mg (28%); pale yellow solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.78 (s, 3H), 5.19 (s, 2H), 7.12 (s, 1H), 7.21-7.26 (m, 2H), 7.54-7.60 (m, 2H), 7.85 (s, 2H), 8.05 (s, 1H). HRMS (ESI$^+$) m/z for $C_{16}H_{14}N_2O_3FS$ ([M+H]$^+$): calculated 333.0704, found 333.0698.

S5. ^{1}H and ^{13}C NMR spectra of the representative compounds

Figure S8. ^{1}H NMR spectra of methyl 2-aminobenzo[*d*]thiazole-6-carboxylate (**A**).

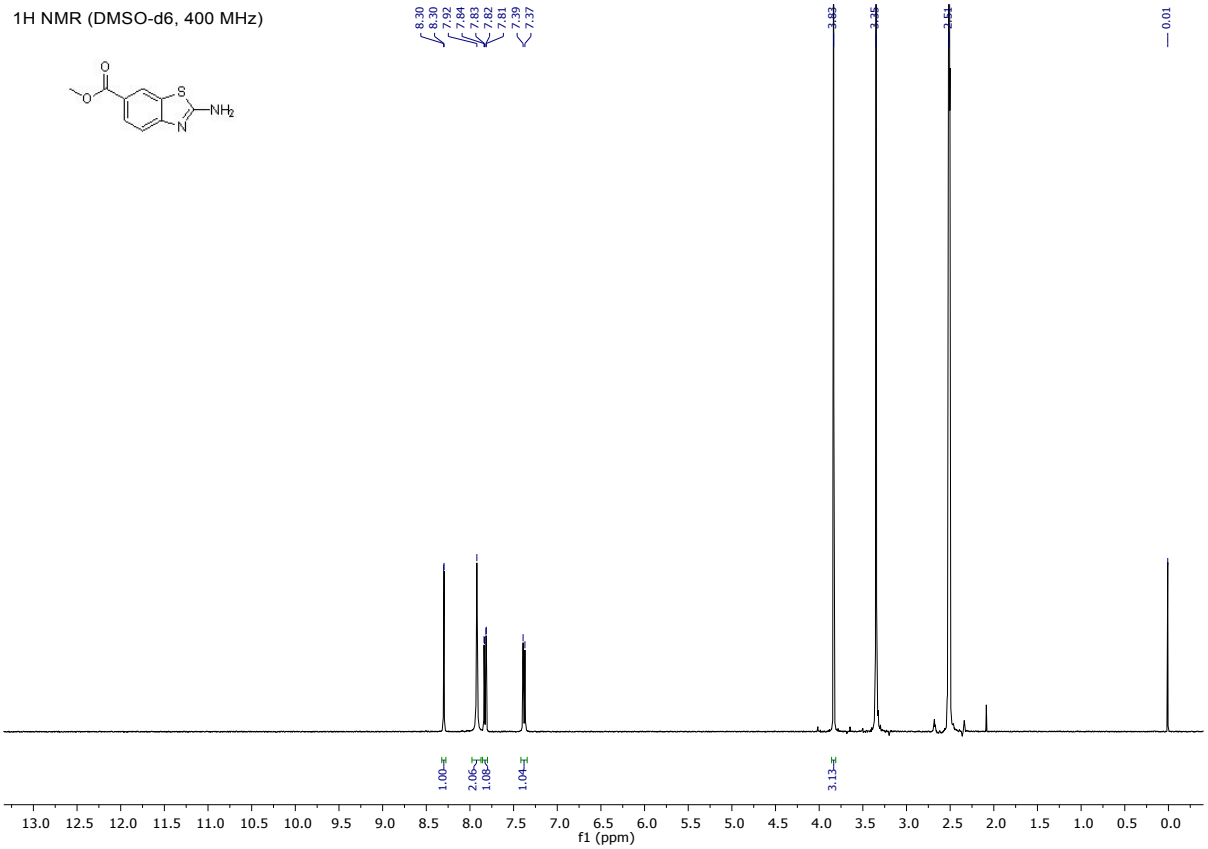


Figure S9. ^{13}C NMR spectra of methyl 2-aminobenzo[*d*]thiazole-6-carboxylate (**A**).

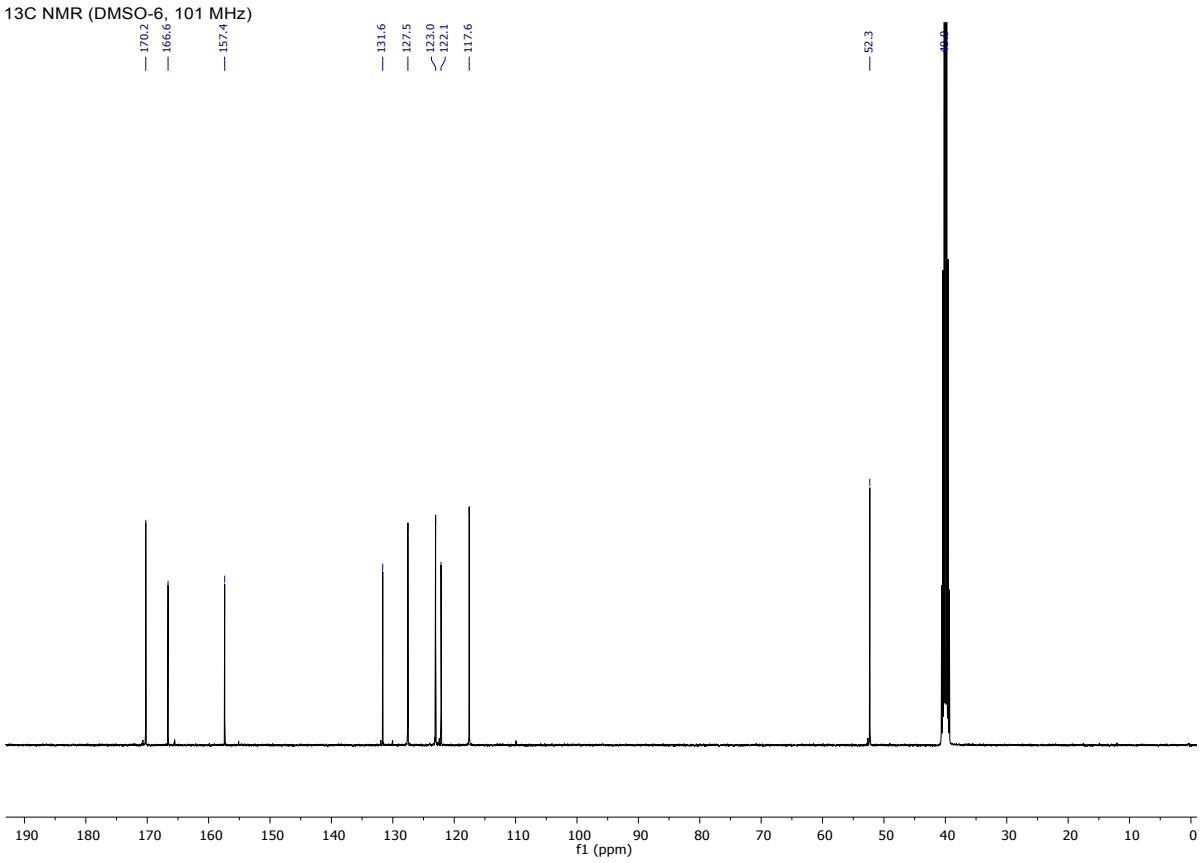


Figure S10. ^{1}H NMR spectra of methyl 2-amino-4-methoxybenzo[*d*]thiazole-6-carboxylate (**5a**).

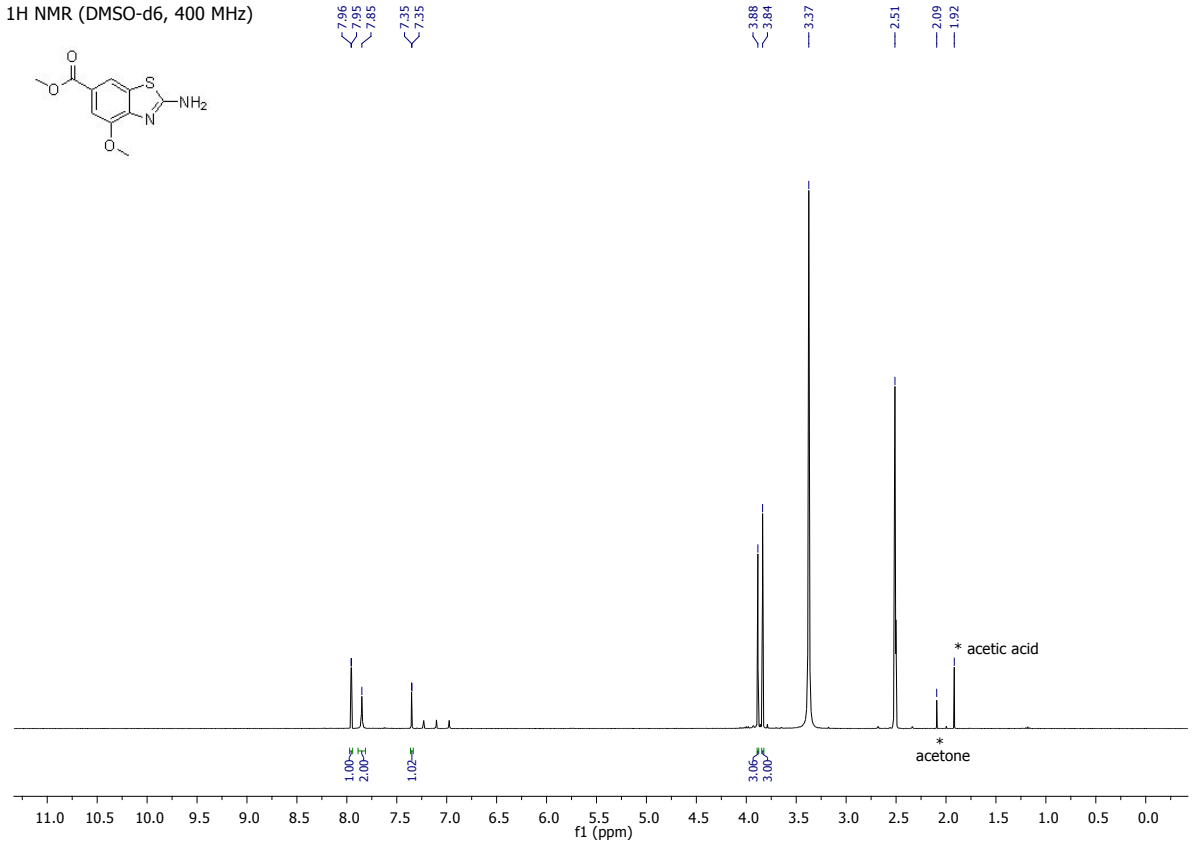


Figure S11. ^{13}C NMR spectra of methyl 2-amino-4-methoxybenzo[*d*]thiazole-6-carboxylate (**5a**).

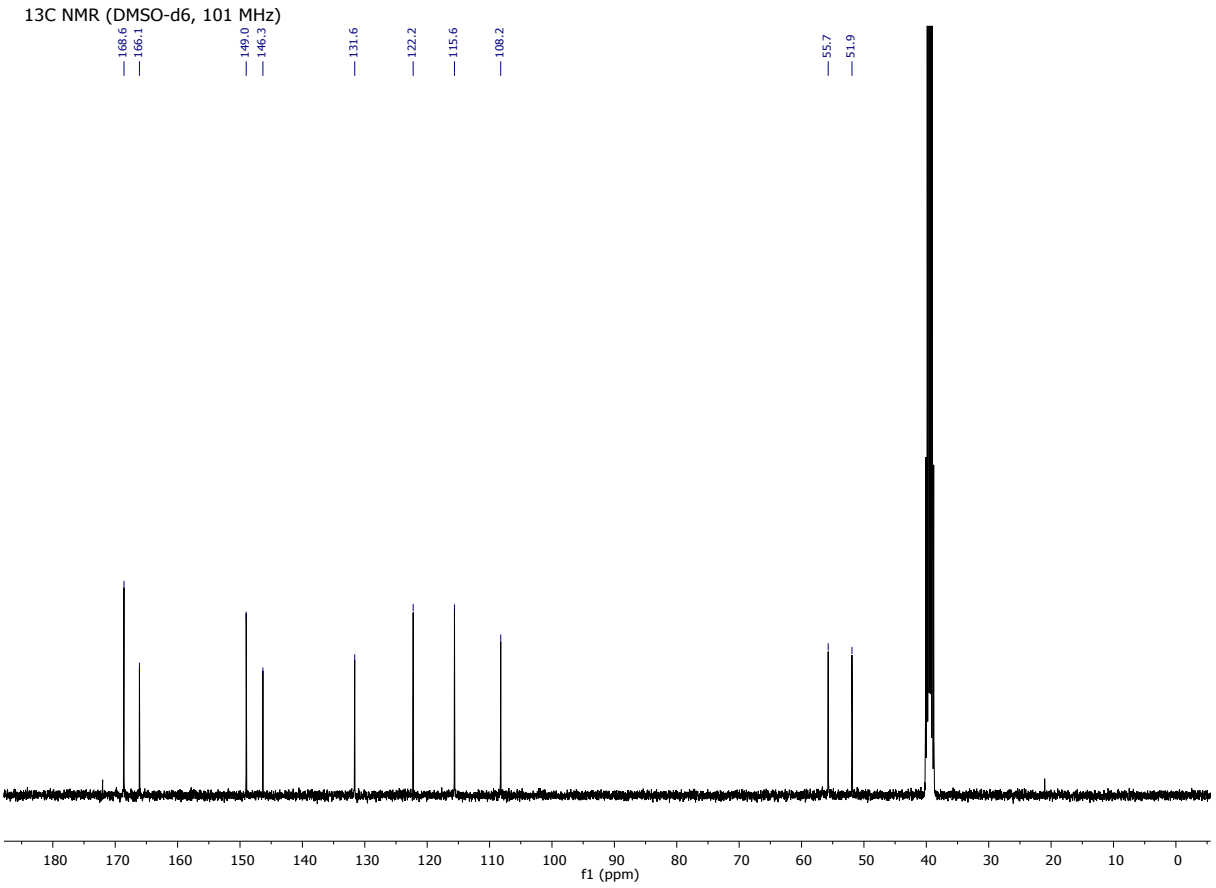


Figure S12. ^{1}H NMR spectra of methyl 2-amino-4-(benzyloxy)benzo[*d*]thiazole-6-carboxylate (**5b**).

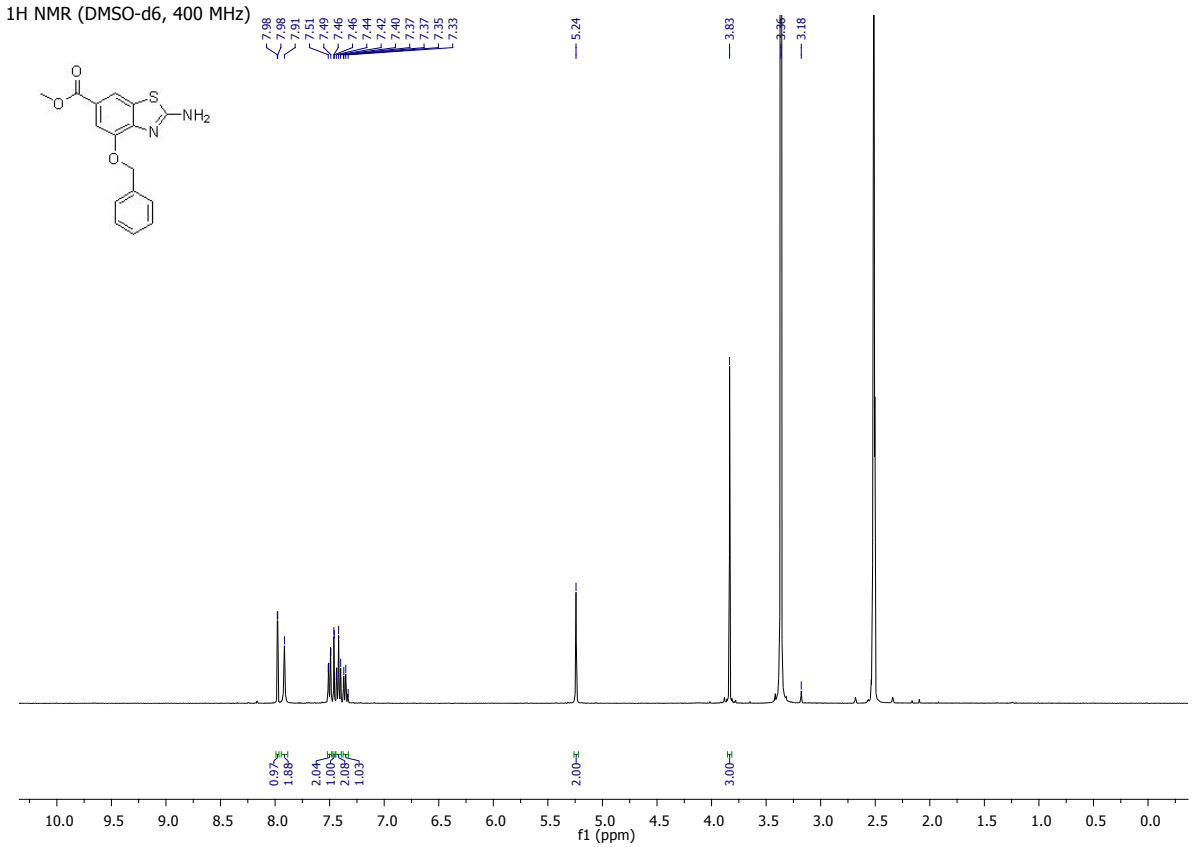


Figure S13. ^{13}C NMR spectra of methyl 2-amino-4-(benzyloxy)benzo[*d*]thiazole-6-carboxylate (**5b**).

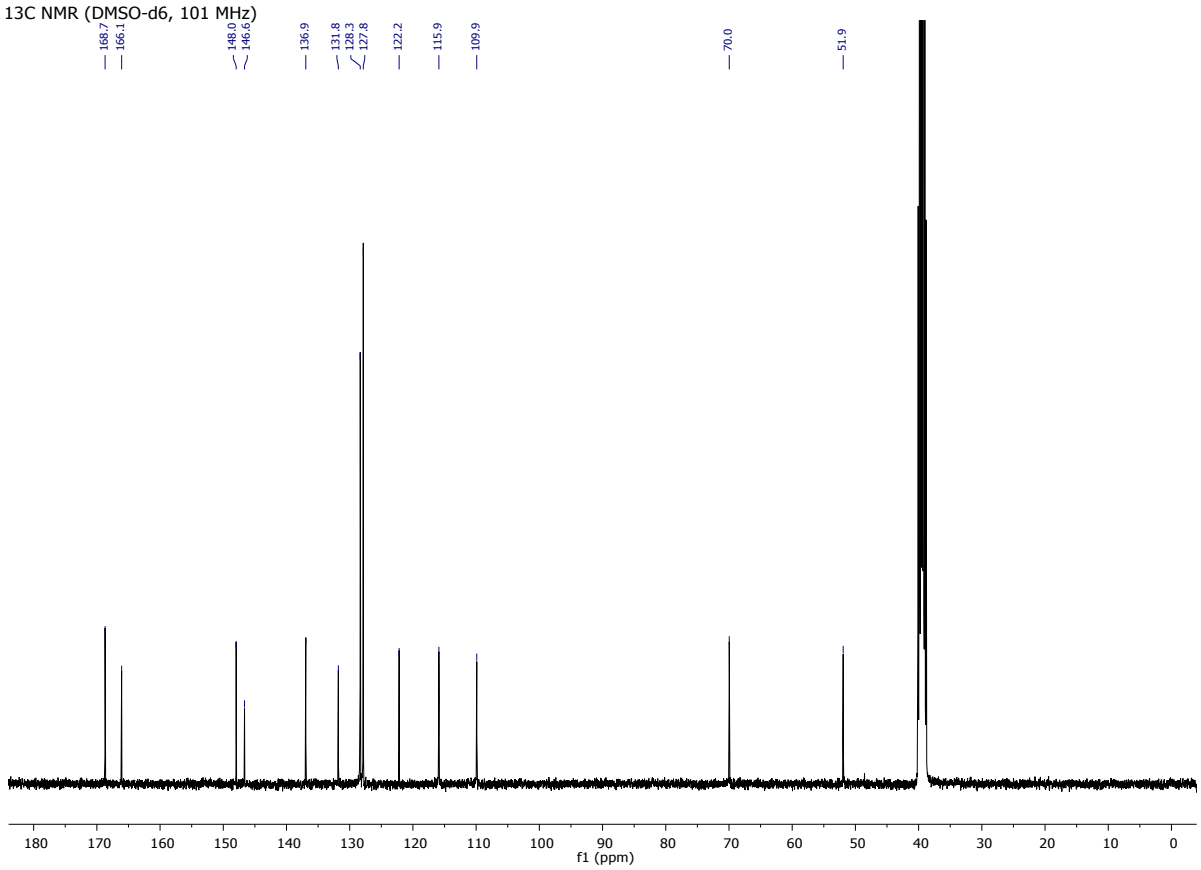


Figure S14. ^{1}H NMR spectra of methyl 2-amino-4-(2-morpholinoethoxy)benzo[*d*]thiazole-6-carboxylate (**5c**).

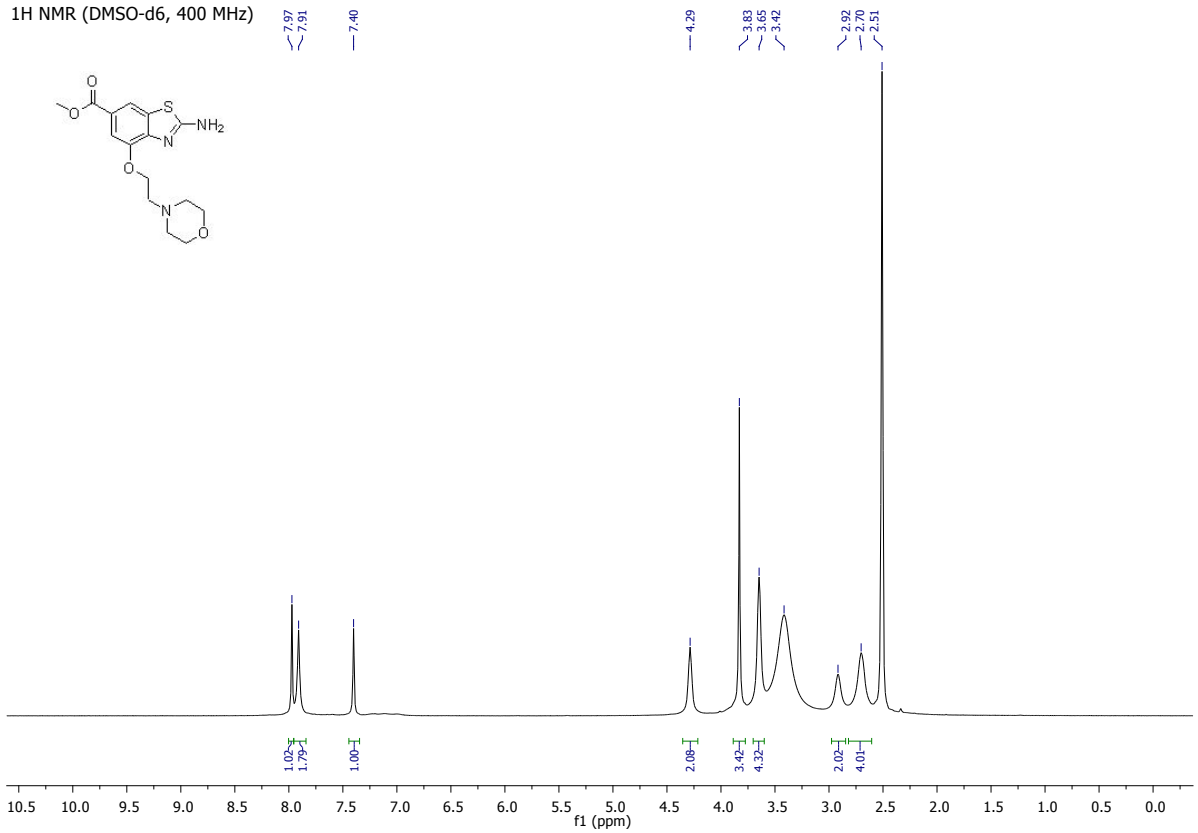


Figure S15. ^{13}C NMR spectra of methyl 2-amino-4-(2-morpholinoethoxy)benzo[*d*]thiazole-6-carboxylate (**5c**).

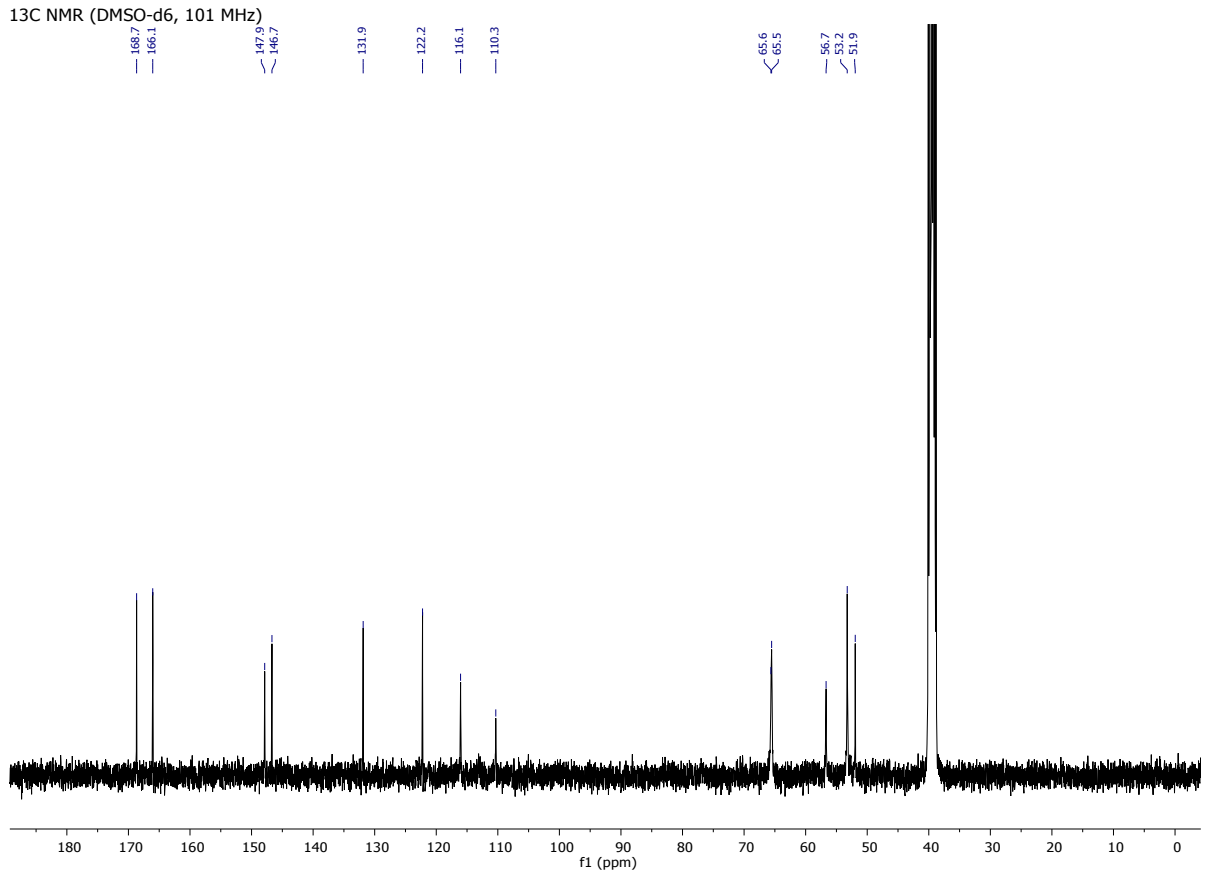


Figure S16. ^{1}H NMR spectra of methyl 2-amino-4-(2-((*tert*-butoxycarbonyl)amino)ethoxy)benzo[*d*]thiazole-6-carboxylate (**5d**).

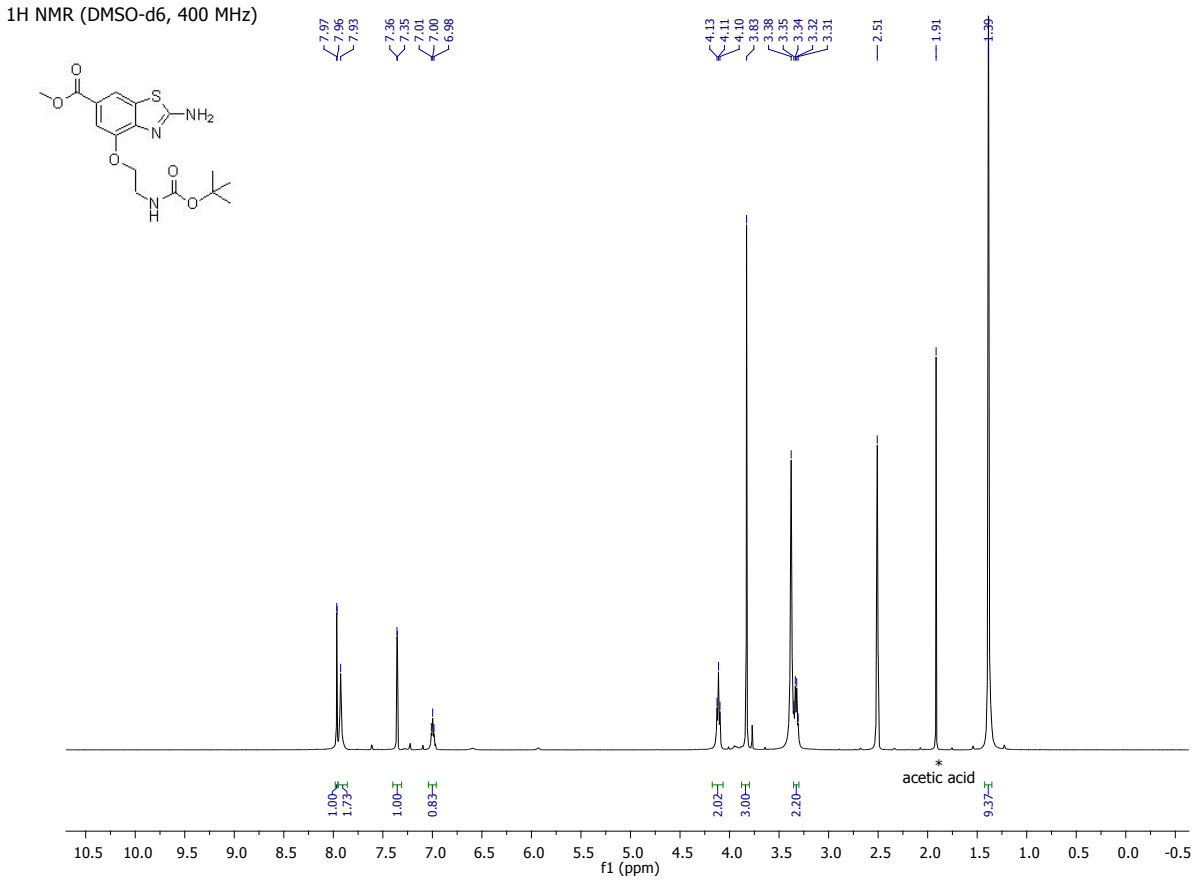


Figure S17. ^{13}C NMR spectra of methyl 2-amino-4-(2-((*tert*-butoxycarbonyl)amino)ethoxy)benzo[*d*]thiazole-6-carboxylate (**5d**).

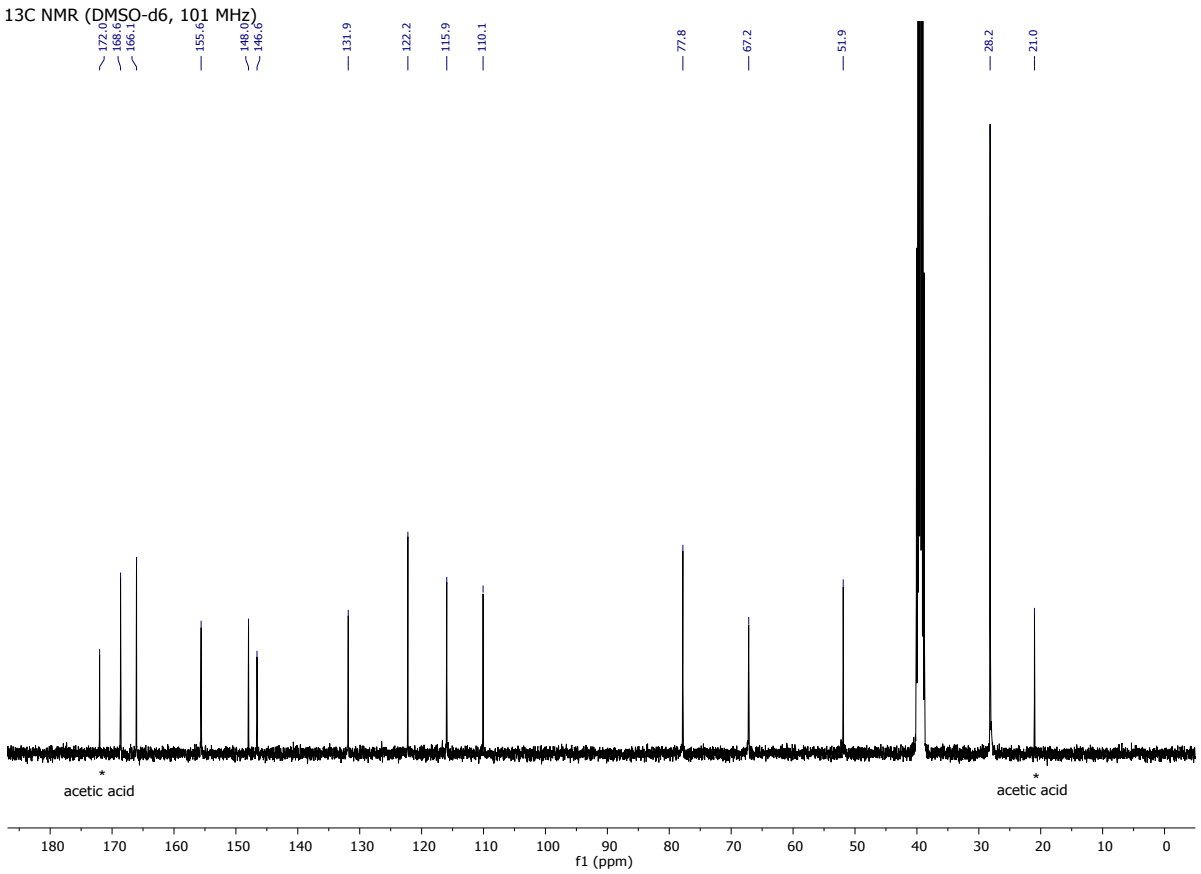


Figure S18. ^{1}H NMR spectra of methyl 2-amino-4-((3-(methoxycarbonyl)benzyl)oxy)benzo[*d*]thiazole-6-carboxylate (**5e**).

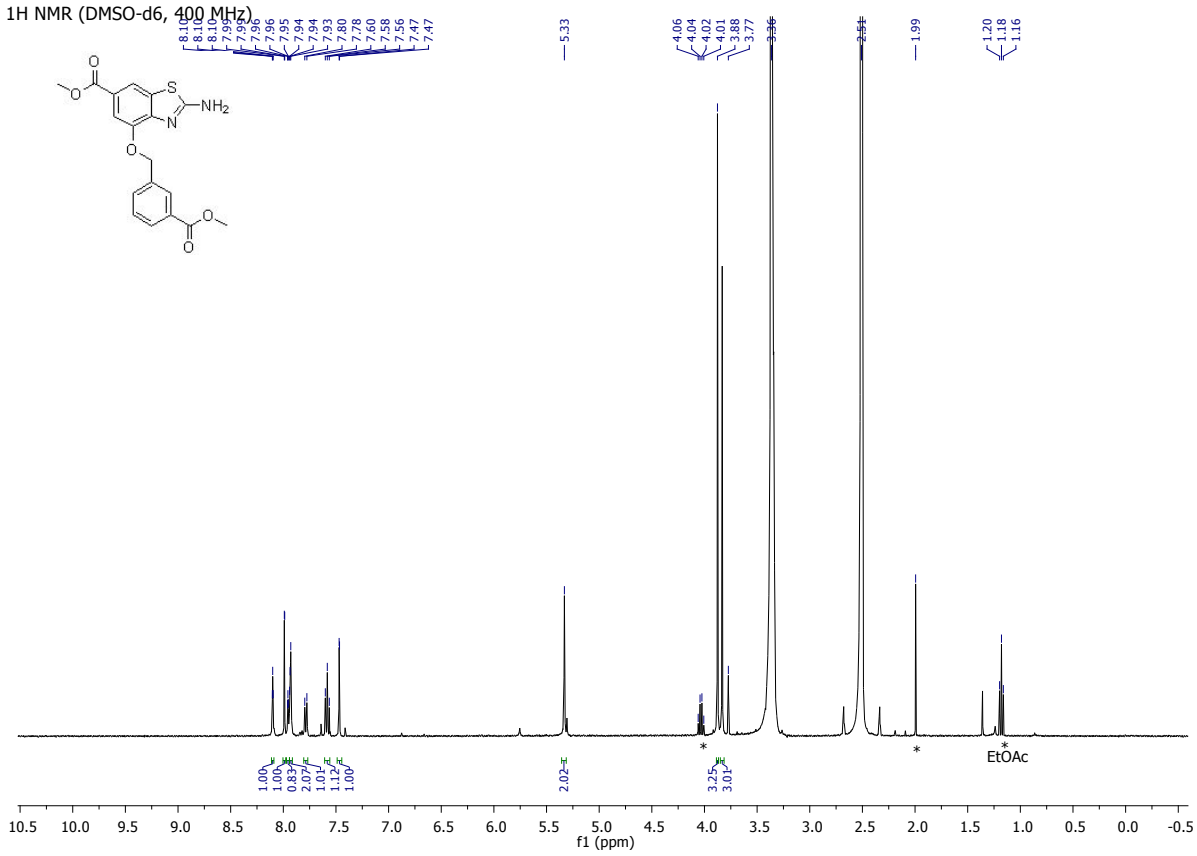


Figure S19. ^{13}C NMR spectra of methyl 2-amino-4-((3-(methoxycarbonyl)benzyl)oxy)benzo[*d*]thiazole-6-carboxylate (**5e**).

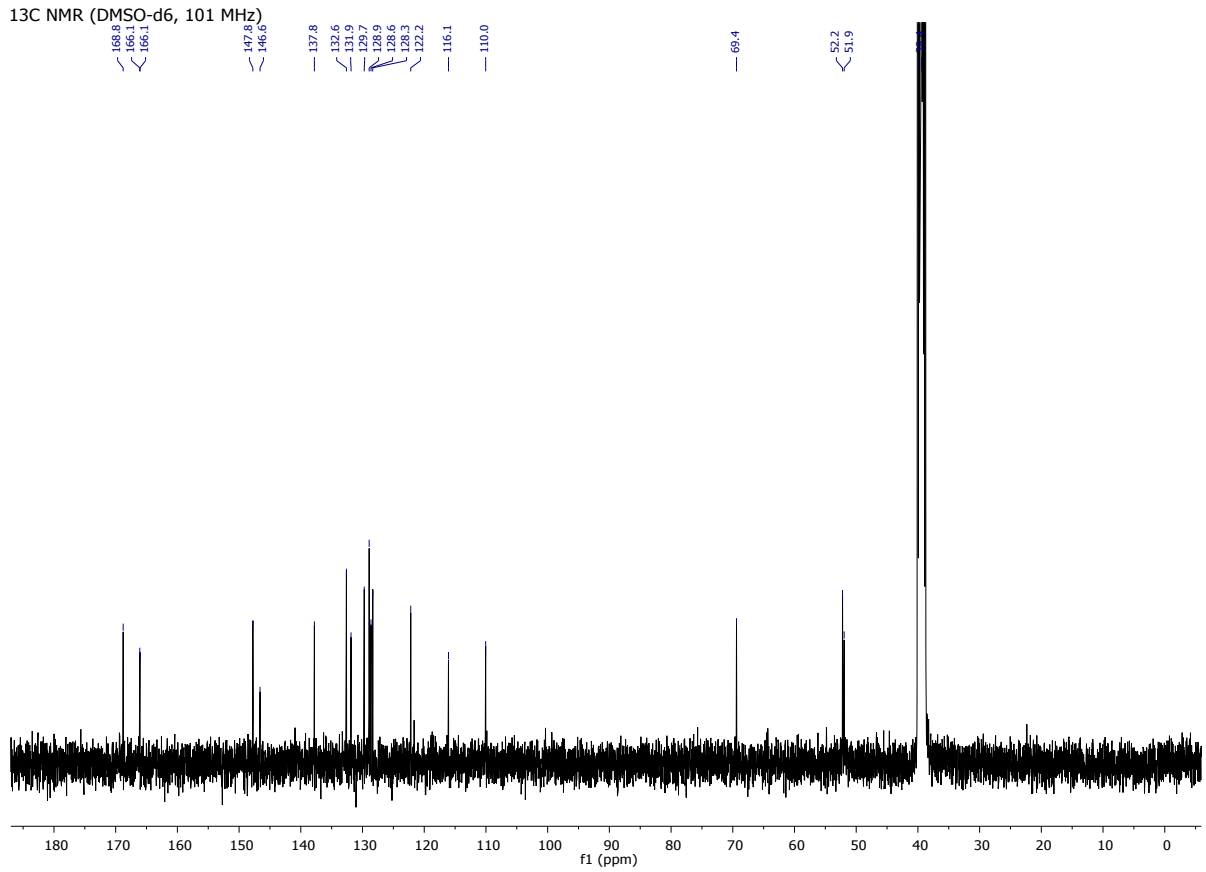


Figure S20. ^{1}H NMR spectra of methyl 2-amino-4-(2-methoxyethoxy)benzo[*d*]thiazole-6-carboxylate (**5f**).

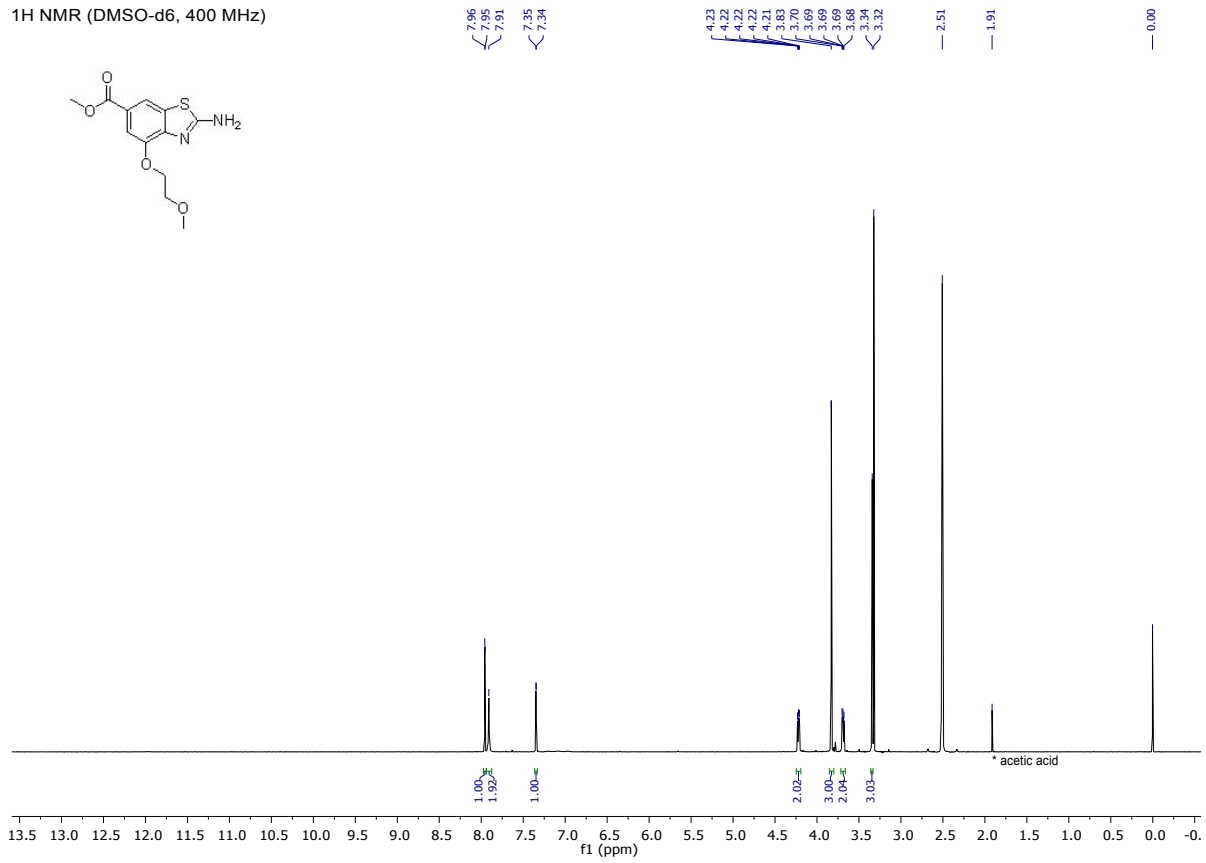


Figure S21. ^{13}C NMR spectra of methyl 2-amino-4-(2-methoxyethoxy)benzo[*d*]thiazole-6-carboxylate (**5f**).

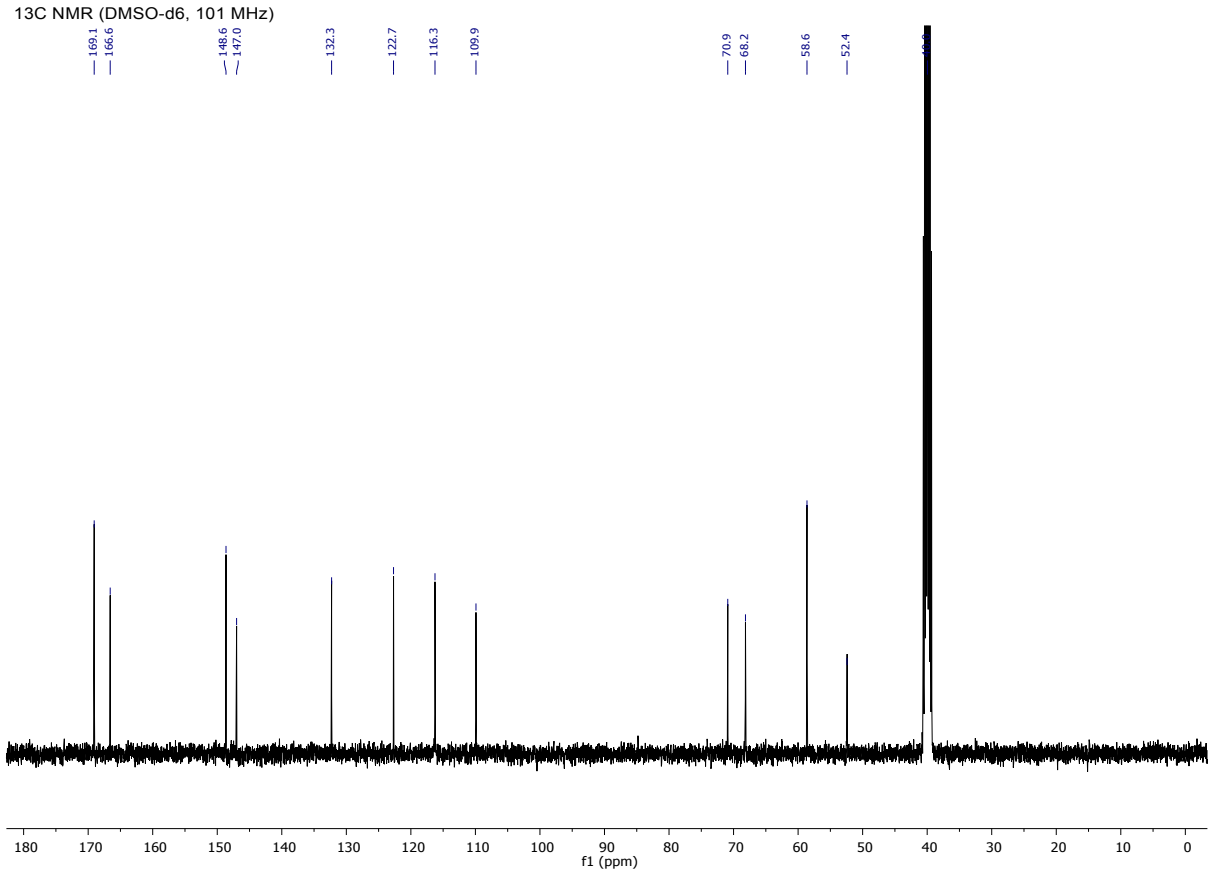


Figure S22. ^{1}H NMR spectra of methyl 2-amino-5-methoxybenzo[*d*]thiazole-6-carboxylate (**11a**).

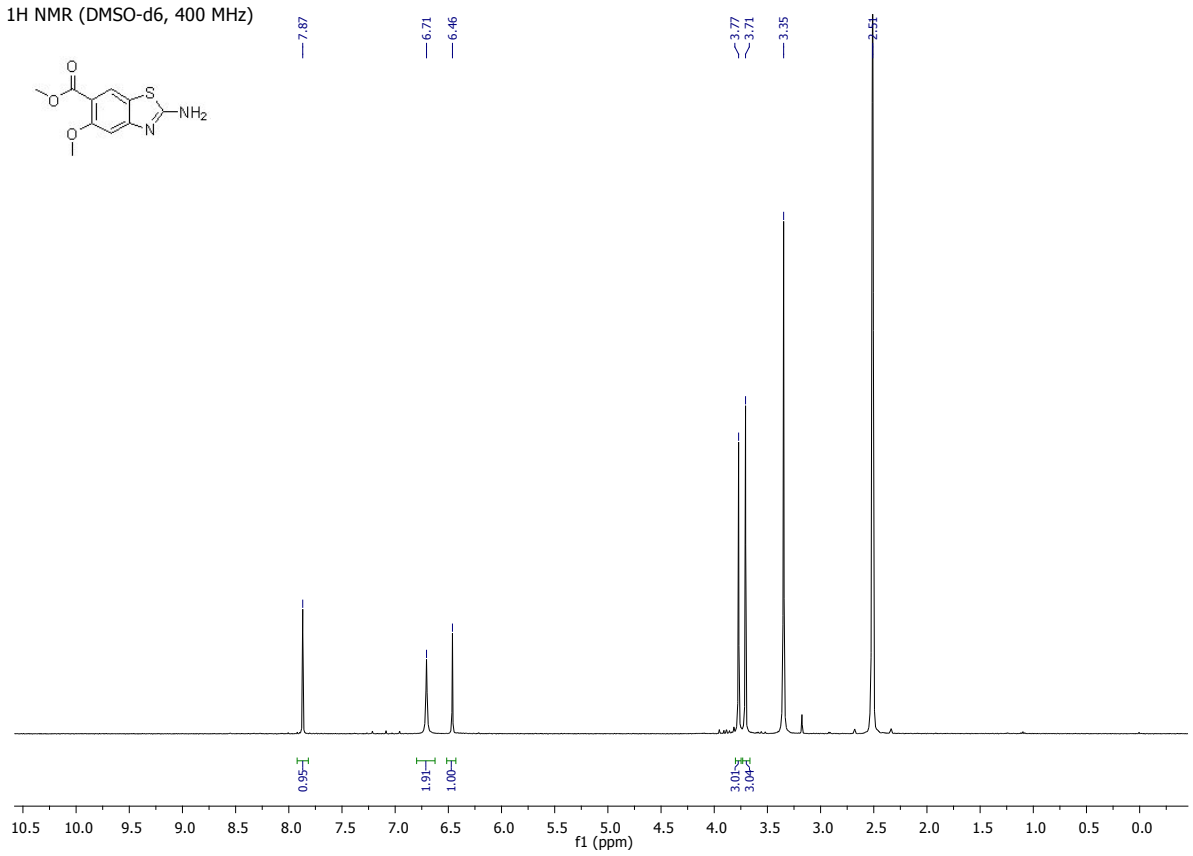


Figure S23. ^{13}C NMR spectra of methyl 2-amino-5-methoxybenzo[*d*]thiazole-6-carboxylate (**11a**).

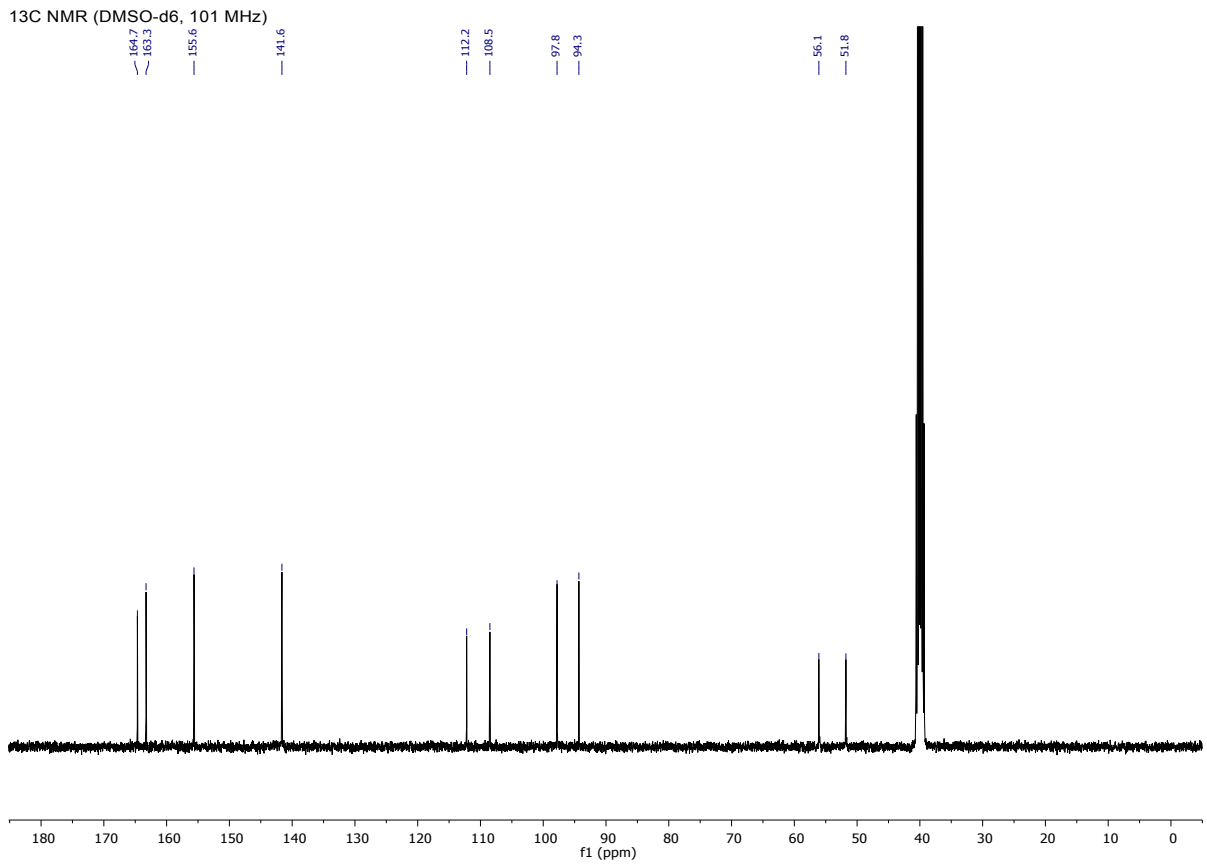


Figure S24. ^{1}H NMR spectra of methyl 2-amino-5-(benzyloxy)benzo[*d*]thiazole-6-carboxylate (**11b**).

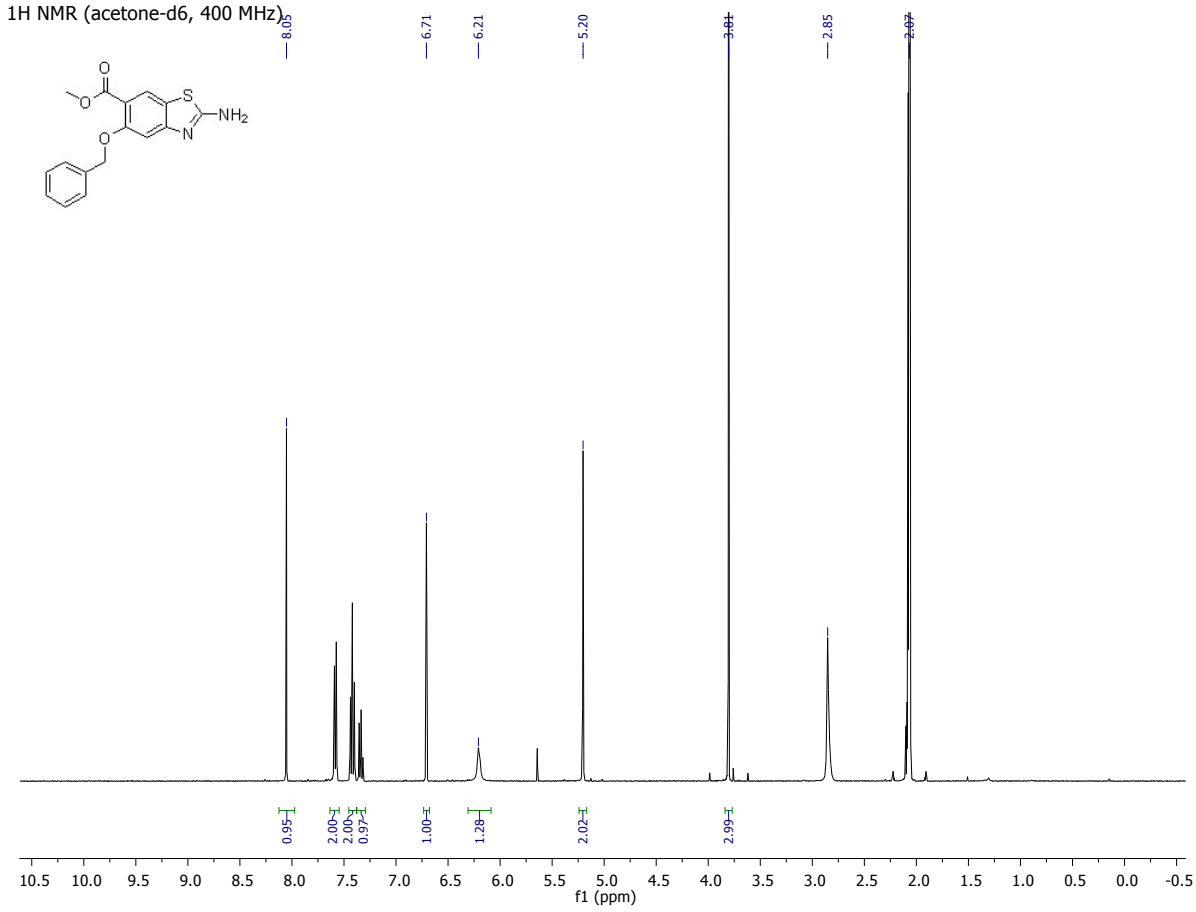


Figure S25. ^{13}C NMR spectra of methyl 2-amino-5-(benzyloxy)benzo[*d*]thiazole-6-carboxylate (**11b**).

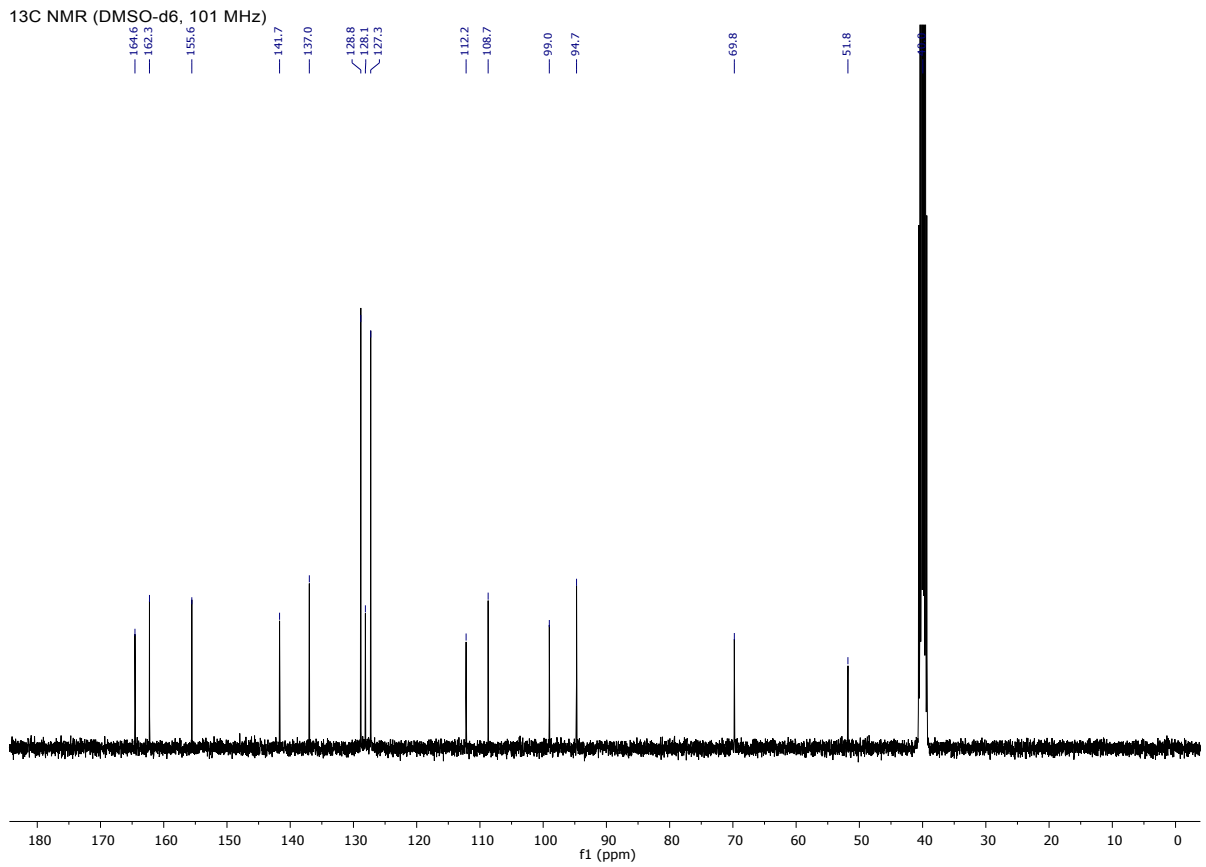


Figure S26. ^{1}H NMR spectra of methyl 2-amino-4-((*tert*-butyldimethylsilyl)oxy)benzo[*d*]thiazole-6-carboxylate (**14**).

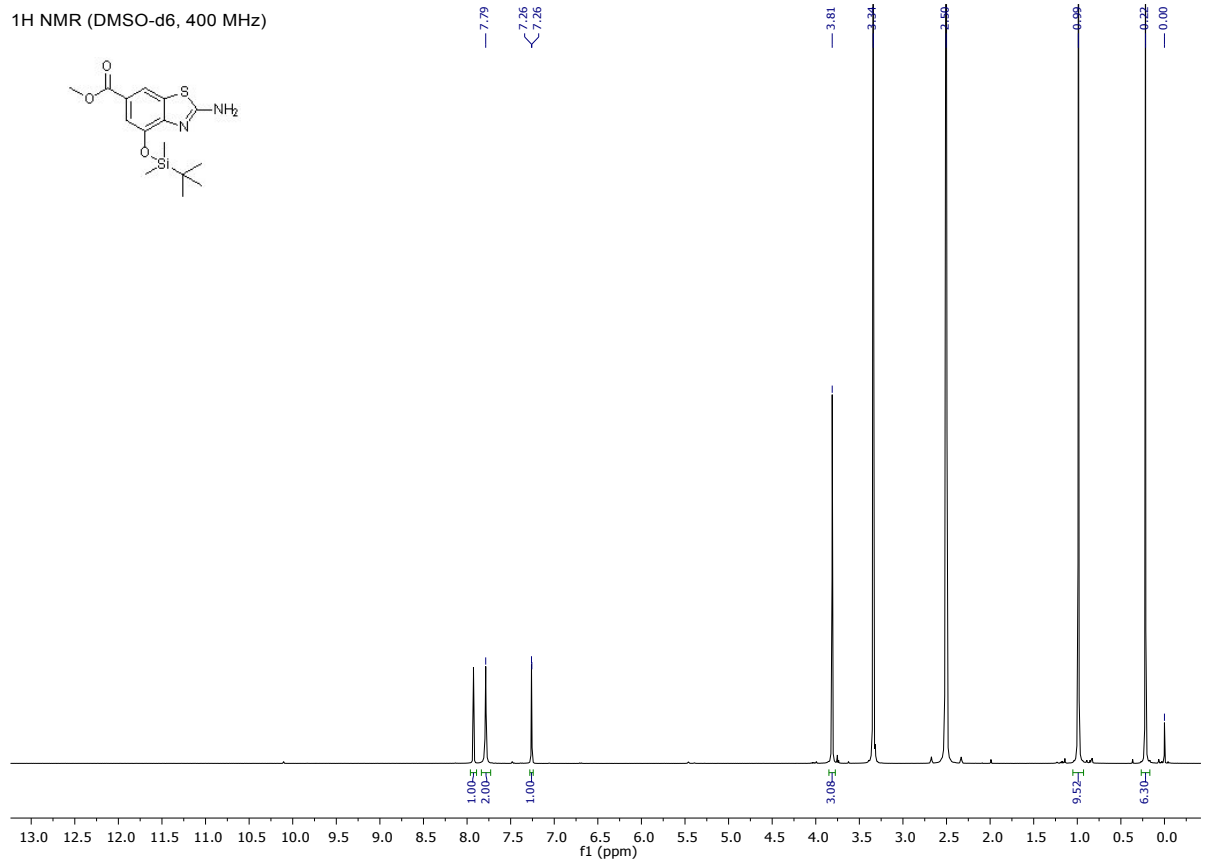


Figure S27. ^{13}C NMR spectra of methyl 2-amino-4-((*tert*-butyldimethylsilyl)oxy)benzo[*d*]thiazole-6-carboxylate (**14**).

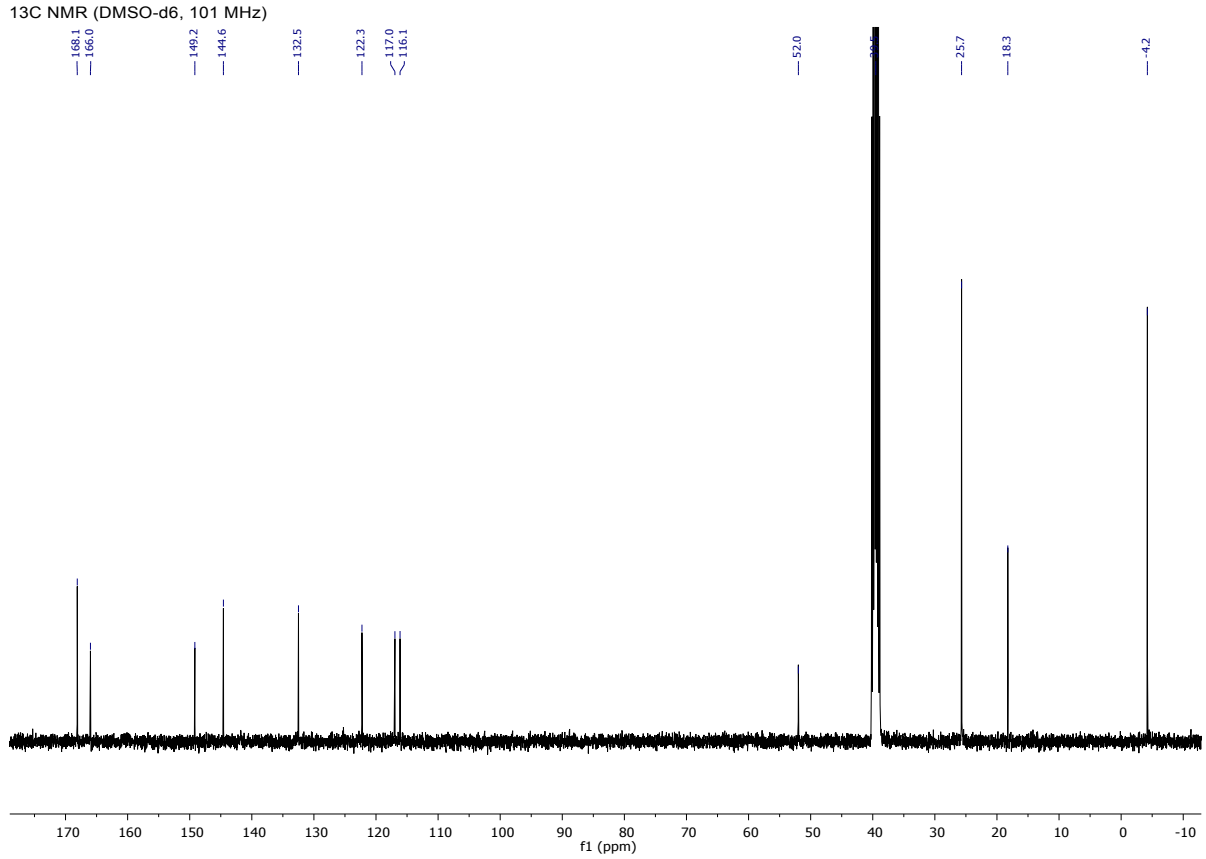


Figure S28. ^{1}H NMR spectra of methyl 2-amino-4-hydroxybenzo[*d*]thiazole-6-carboxylate (**15**).

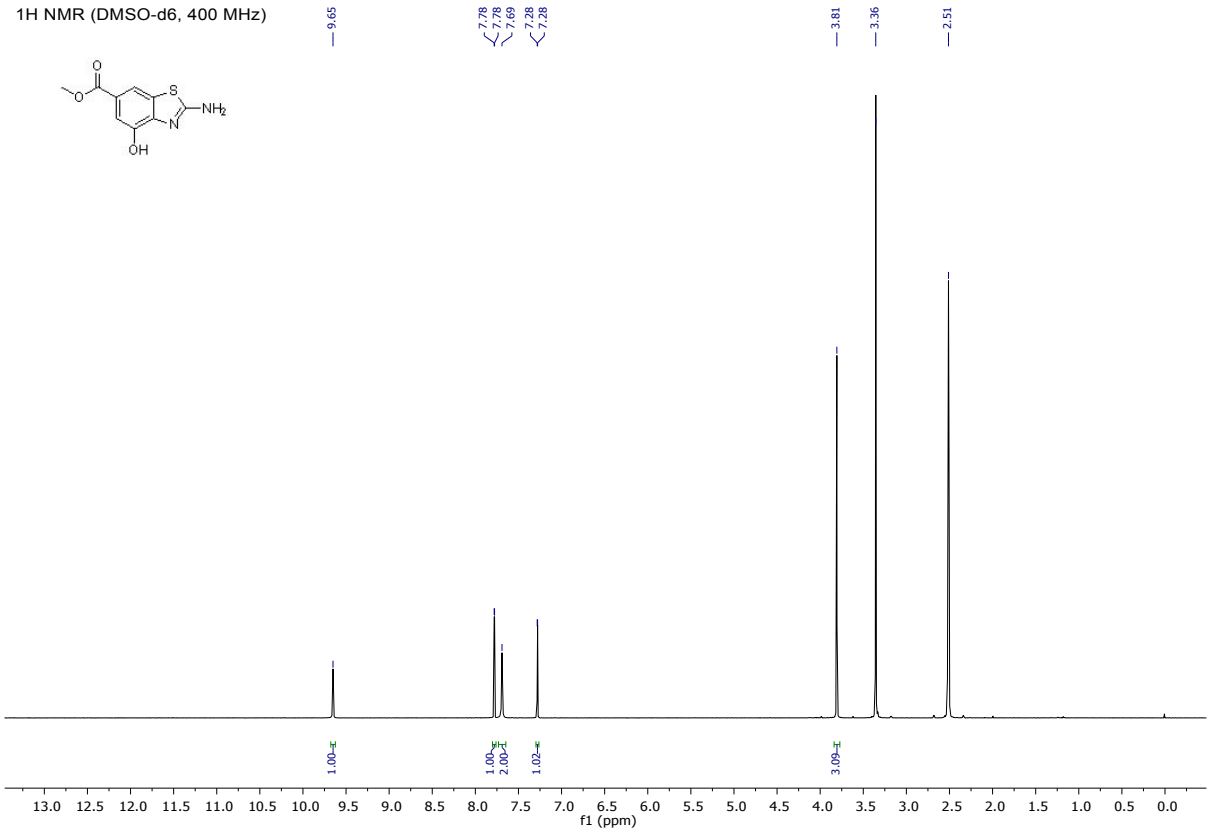


Figure S29. ^{13}C NMR spectra of methyl 2-amino-4-hydroxybenzo[*d*]thiazole-6-carboxylate (**15**).

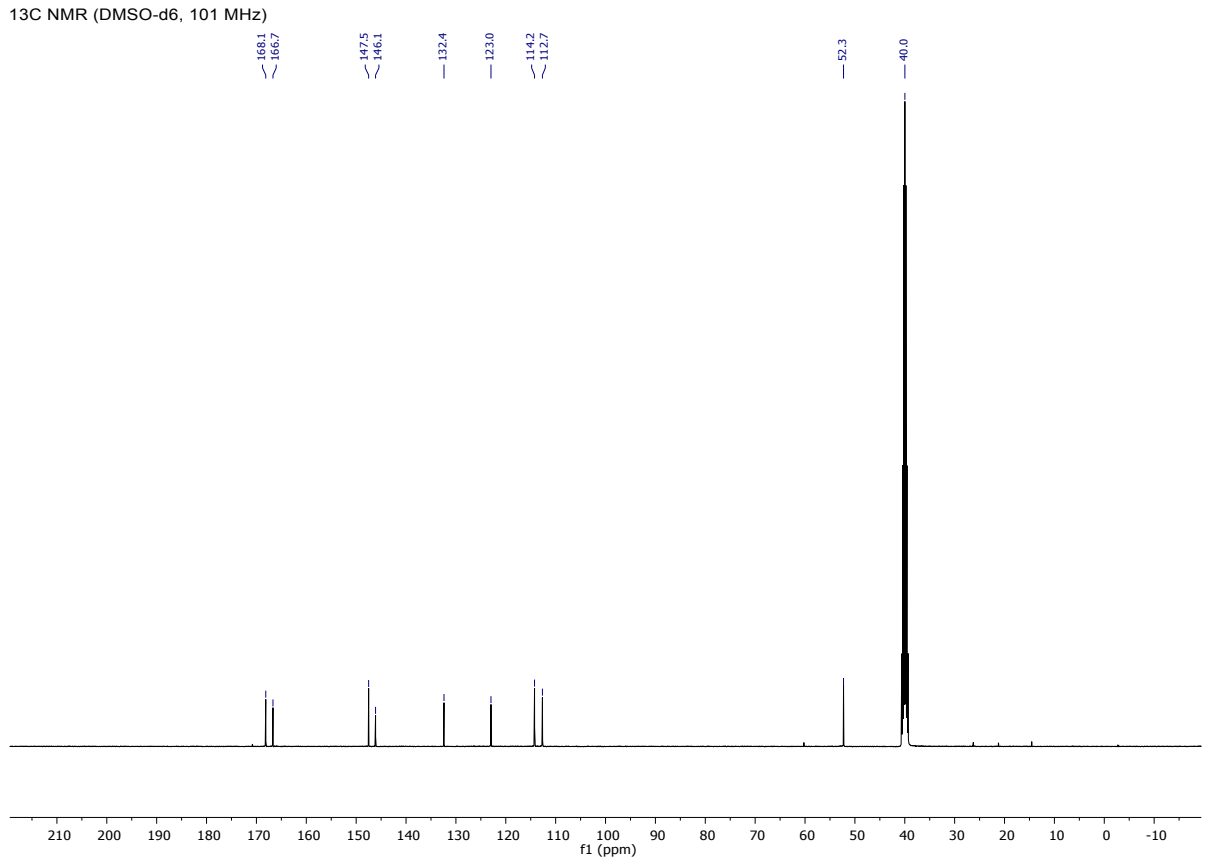


Figure S30. ^{1}H NMR spectra of methyl 2-amino-5-((*tert*-butyldimethylsilyl)oxy)benzo[*d*]thiazole-6-carboxylate (**17**).

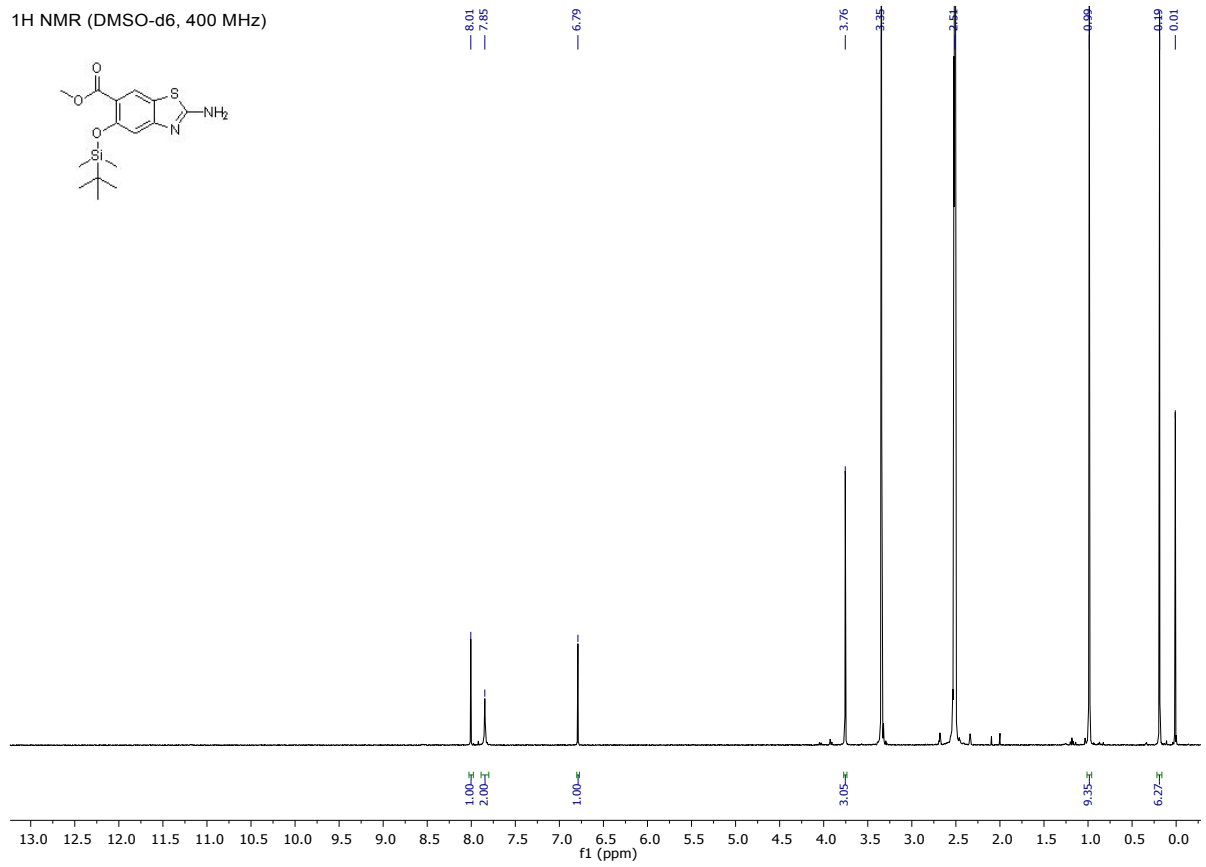


Figure S31. ^{13}C NMR spectra of methyl 2-amino-5-((*tert*-butyldimethylsilyl)oxy)benzo[*d*]thiazole-6-carboxylate (**17**).

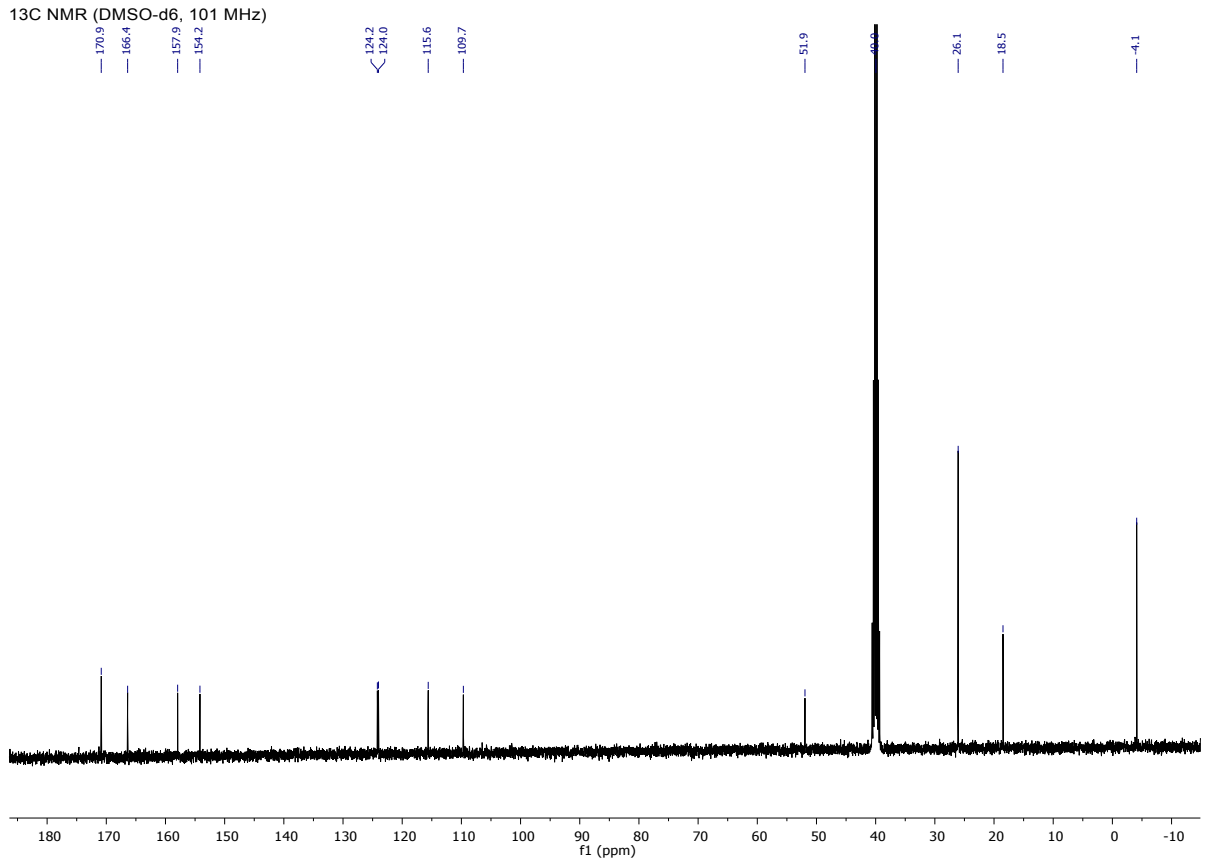


Figure S32. ^{1}H NMR spectra of methyl 2-amino-5-hydroxybenzo[*d*]thiazole-6-carboxylate (**18**).

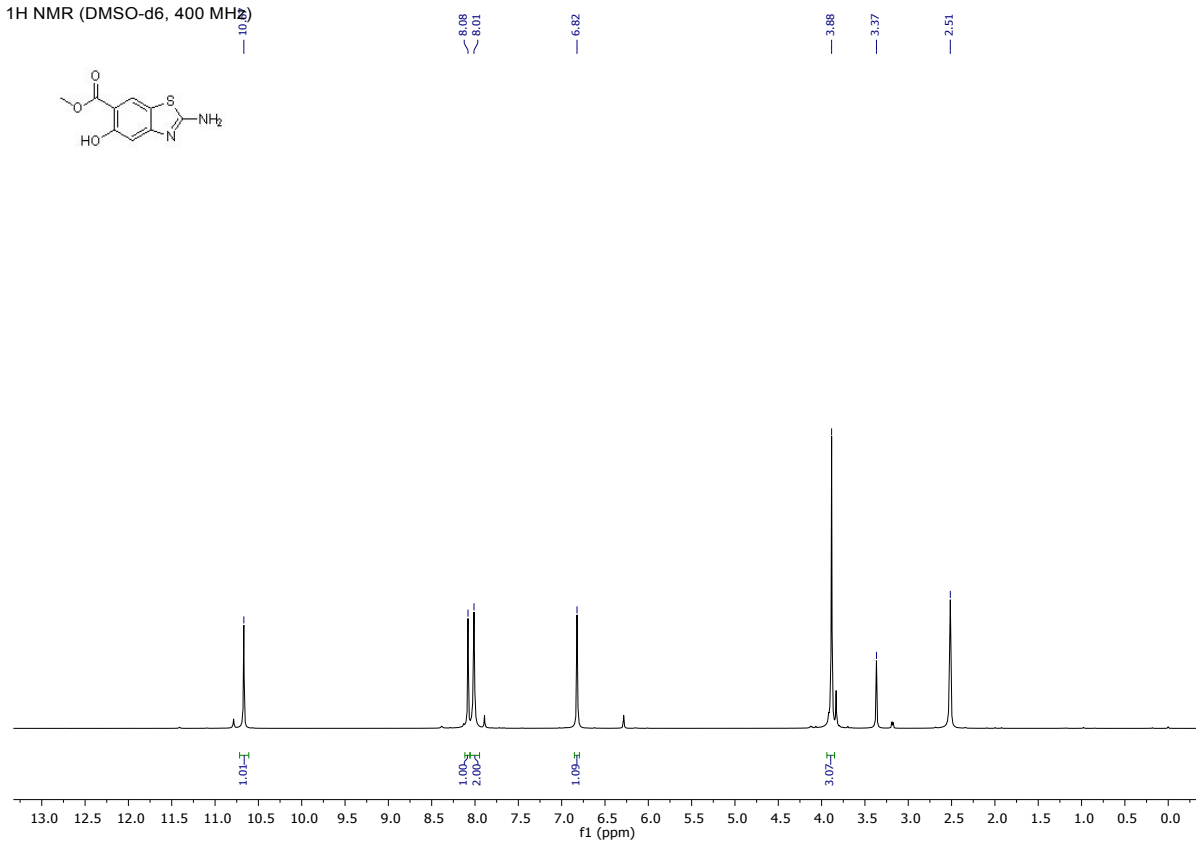


Figure S33. ^{13}C NMR spectra of methyl 2-amino-5-hydroxybenzo[*d*]thiazole-6-carboxylate (**18**).

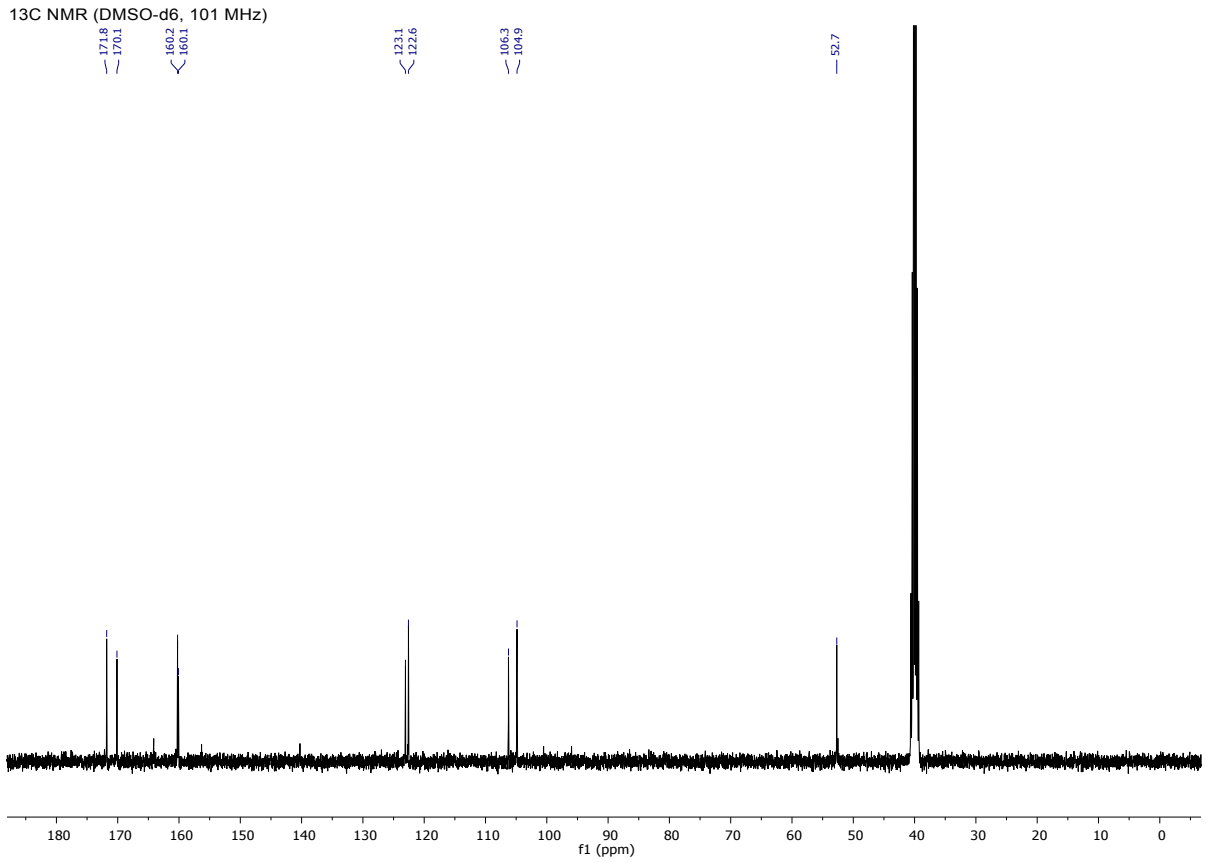


Figure S34. ^{1}H NMR spectra of methyl 2-amino-4-((4-fluorobenzyl)oxy)benzo[*d*]thiazole-6-carboxylate (**19a**).

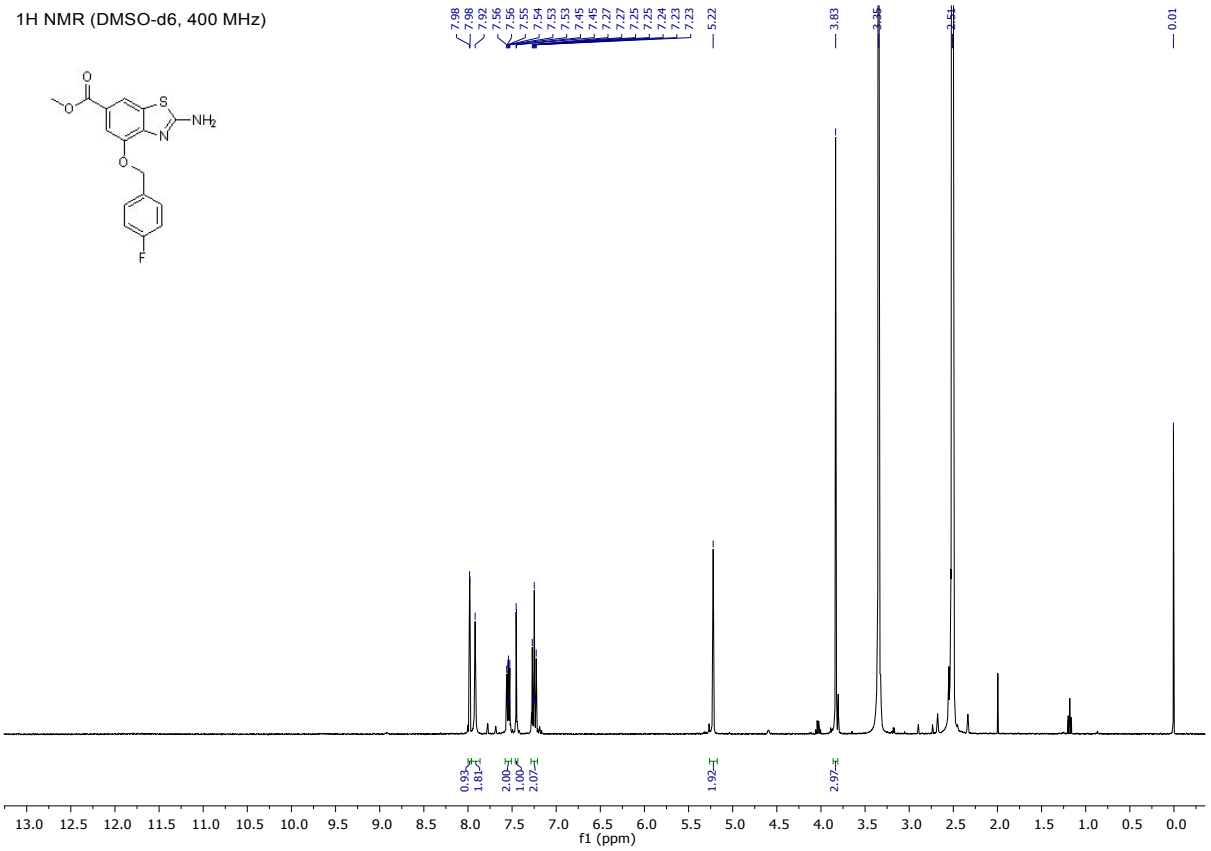


Figure S35. ^{13}C NMR spectra of methyl 2-amino-4-((4-fluorobenzyl)oxy)benzo[*d*]thiazole-6-carboxylate (**19a**).

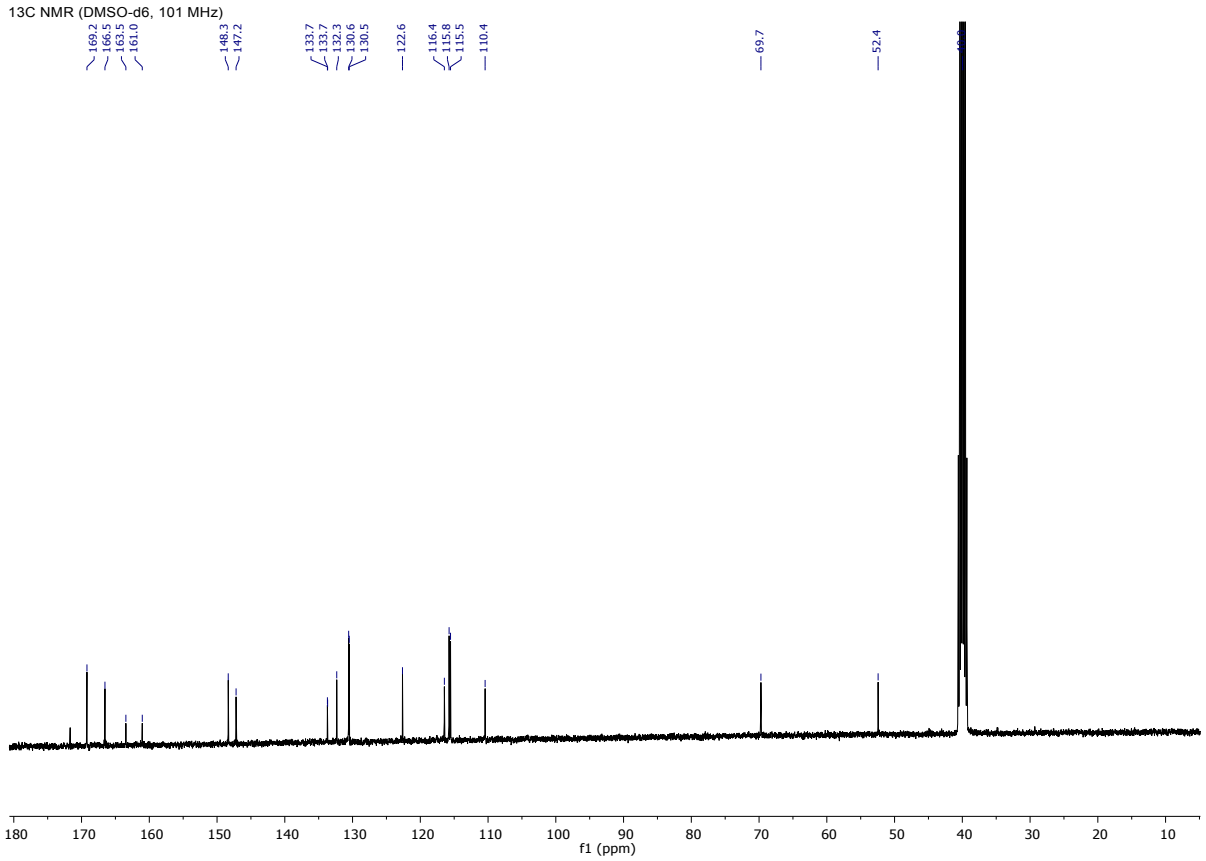


Figure S36. ^{1}H NMR spectra of methyl 2-amino-4-((4-(methoxycarbonyl)benzyl)oxy)benzo[*d*]thiazole-6-carboxylate (**19c**).

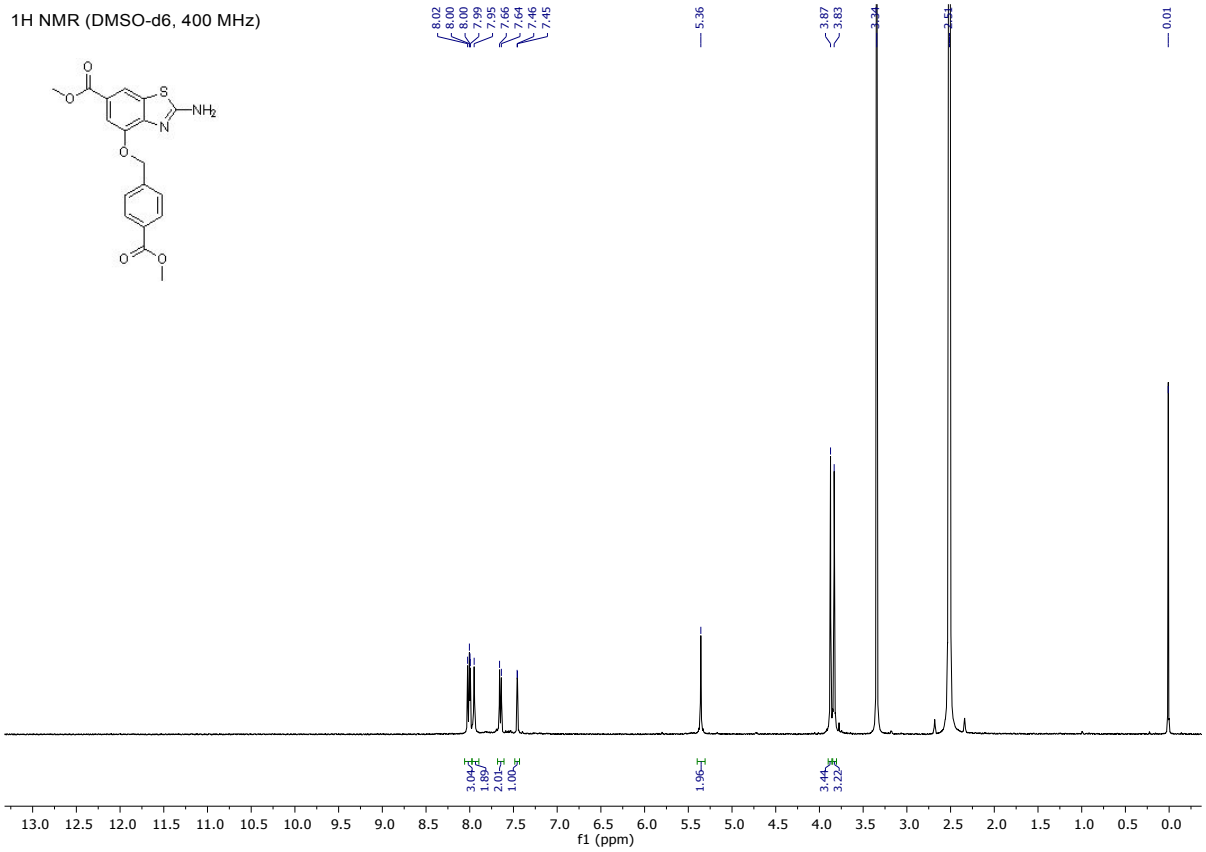
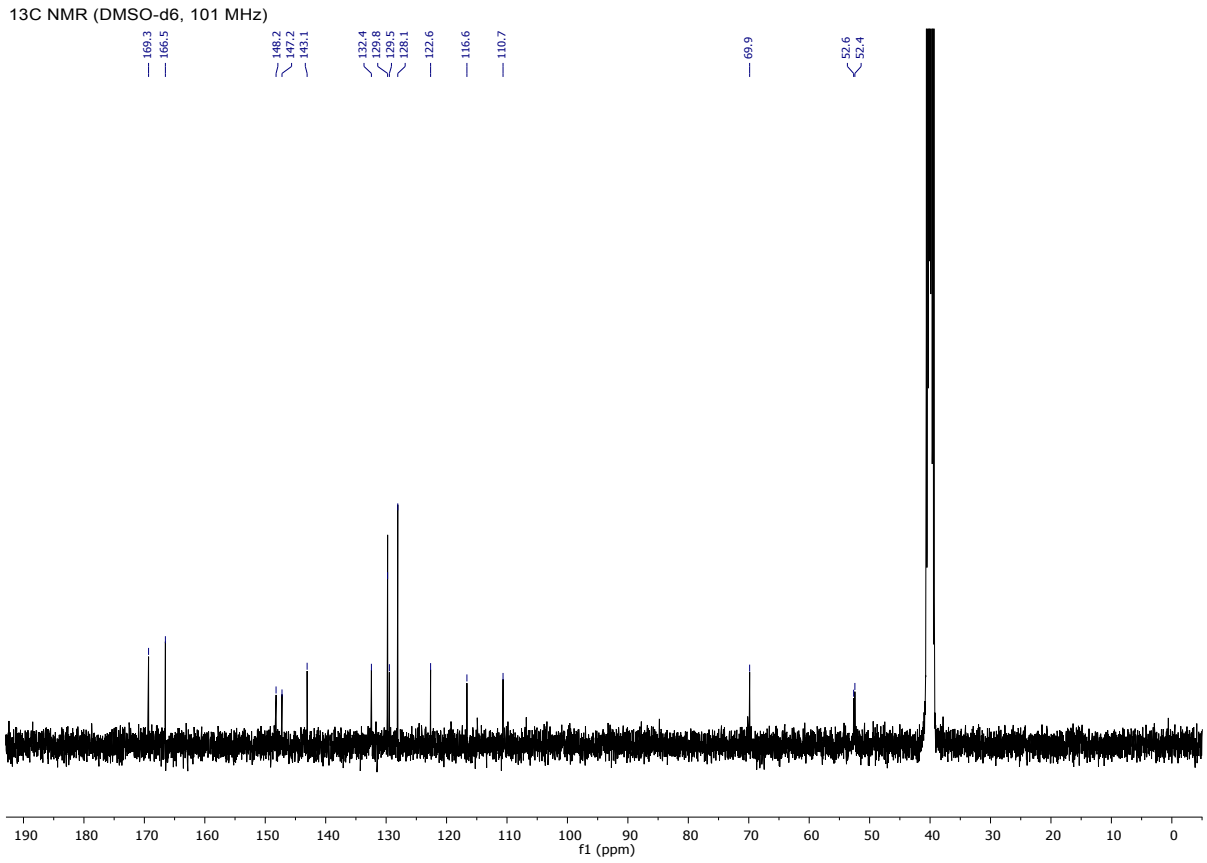


Figure S37. ^{13}C NMR spectra of methyl 2-amino-4-((4-(methoxycarbonyl)benzyl)oxy)benzo[*d*]thiazole-6-carboxylate (**19c**).



S6. References

(1) Charrier, N.; Demont, E.; Dunsdon, R.; Maile, G.; Naylor, A.; O'Brien, A.; Redshaw, S.; Theobald, P.; Vesey, D.; Walter, D. Synthesis of Indoles: Efficient Functionalisation of the 7-Position. *Synthesis.* **2006**, *2006*, 3467–3477.

(2) Durcik, M.; Lovison, D.; Skok, Ž.; Durante Cruz, C.; Tammela, P.; Tomašič, T.; Benedetto Tiz, D.; Draskovits, G.; Nyerges, Á.; Pál, C.; Ilaš, J.; Peterlin Mašič, L.; Kikelj, D.; Zidar, N. New *N*-phenylpyrrolamide DNA gyrase B inhibitors: Optimization of efficacy and antibacterial activity. *Eur. J. Med. Chem*. **2018**, *154*, 117–132.

(3) Murphy, N.-S.; Prabhakaran, P.; Azzarito, V.; Plante, J.-P.; Hardie, M.-J.; Kilner, C.-A.; Warriner, S.-L.; Wilson, A.-J. Solid-Phase Methodology for Synthesis ofO-Alkylated Aromatic OligoamideInhibitors ofa-Helix-Mediated Protein–Protein Interactions. *Chem. Eur. J.* **2013**, *19*, 5546-5550.

(4) Toyota, T.; Yoshinaga, E. JP Patent 2011051979 A, **2011**.

(5) Tully, D.-C.; Rucker, P.-V.; Chianelli, D.; Williams, J.; Vidal, A.; Alper, P.-B.; Mutnick, D.; Bursulaya, B.; Schmeits, J.; Wu, X.; Bao, D.; Zoll, J.; Kim, Y.; Groessi, T.; McNamara, P.; Seidel, H.-M.; Molteni, V.; Liu, B.; Phimister, A.; Joseph, S.-B.; Laffitte, B. Discovery of Tropifexor (LJN452), a Highly Potent Non-bile Acid FXR Agonist for the Treatment of Cholestatic Liver Diseases and Nonalcoholic Steatohepatitis (NASH). *J. Med. Chem.* **2017**, *60*, 9960-9973.

(6) Chen, J.; Lu, W.; Chen, H.; Bian, X.; Yang, G. A New Series of Salicylic Acid Derivatives as Non-saccharide α-Glucosidase Inhibitors and Antioxidants. *Biol. Pharm. Bull*. **2019**, *42*, 231-246.

(7) Zhang L, Hu X. Room temperature C(sp2)–H oxidative chlorination via photoredox catalysis *Chem. Sci.* **2017**, *8*, 7009-7013.

(8) Shroot, B.-V.; Eustache, J.; Bernardon, J.-M. EP Patent 232199 A2, **1987**.