Efficient Synthesis of Benzothiazinone Analogues with Activity against Intracellular *Mycobacterium tuberculosis*

Adrian Richter, Gagandeep Narula, Ines Rudolph, Rüdiger W. Seidel, Christoph Wagner, Yossef Av-Gay, and Peter Imming*
**Synthetic materials and methods** All chemicals were purchased from Sigma Aldrich, Alfa Aesar, VWR, Carl Roth, Fisher Scientific or Acros Organics and were used without further purification. All organic solvents, piperidine, 2,6- dimethylpiperidine, TEA (triethylamine), and DIPEA (N,N-diisopropylethylamine) were distilled prior use and stored with molecular sieve 3 Å. All solids were dried in a glass oven (Büchi TO-51, Büchi Labortechnik, Flawil, Switzerland) at 60 °C, 20 mbar for 60-120 min prior to use. The notation hexane in the description of the syntheses refers to n-hexane. Column chromatography was carried out using Merck silica gel 60 (63-200 µm). Flash chromatography was performed on a puriFlash® 430 instrument (Interchim, Montluçon, France). Prepacked columns with silica gel (30 µm) were used. The maximum compound load per column was 5 % (m/m) of the silica gel quantity. NMR spectra were recorded on an Agilent Technologies VNMRS 400 MHz spectrometer. Chemical shifts are reported relative to the residual solvent signal (CDCl₃ δ_H = 7.26 ppm; δ_C = 77.10 ppm; CD₃OD δ_H = 3.31 ppm; DMSO-d₆ δ_H = 2.50 ppm). Abbreviations: s = singlet, bs = broad singlet, d = doublet, dd = doublet of doublets, m = multiplet. ESI mass spectra were measured on a Thermo Finnigan LCQ Classic spectrometer and high-resolution mass spectra (HRMS) were recorded on a Thermo Fisher Scientific LTQ Orbitrap XL mass spectrometer. APCI mass spectra were measured with an Advion expression® compact mass spectrometer. Electron impact (EI) mass spectra were recorded on an AMD 402 of AMD Intectra GmbH. Final compounds were confirmed to be of >95% purity based on HPLC. Purity was measured by UV absorbance at 254 nm. The HPLC consists of an XTerra RP18 column (3.5 µm, 3.9 mm × 100 mm) from the manufacturer Waters (Milford, MA, USA) and two LC-10AD pumps, a SPD-M10A VP PDA detector, and a SIL-HT autosampler, all from the manufacturer Shimadzu (Kyoto, Japan). The mobile phase was a gradient of methanol/ water (starting at 95% water going to 5% water) with 0.05 % TFA.
**General procedure A - Synthesis of aromatic carboxylic acid chlorides** The aromatic carboxylic acid is dissolved in toluene and after addition of SOCl₂ (2 eq.) heated for 2 h under reflux. The solvent is then removed under vacuum and the carboxylic acid chloride produced is used for the next reaction step without further work-up.

**General procedure B - Synthesis of thioureas with thiocarbonyldiimidazole** Thiocarbonyldiimidazole is dissolved in THF in a single-neck flask, the amine is added as solution in THF. The reaction mixture is stirred for 2 h at room temperature, followed by heating to 55 °C for 1 h. After the reaction is complete, 2/3 of the THF volume is removed under vacuum, 2 M ammonia solution in MeOH (2-5 eq. ammonia) is added and the flask is sealed tightly. After 15 h reaction at room temperature, the addition of the 2 M ammonia (2-5 eq. ammonia) solution is repeated and the mixture is heated to 50 °C for 8 h. The solvent is removed under vacuum and the thiourea can be purified by column chromatography or by recrystallisation from isopropanol/diisopropyl ether (1:1).

**General procedure C - Synthesis of benzothiazinones by the thiourea pathway** The thiourea prepared according to general procedure B is added to toluene in a multi-neck flask and the suspension is heated to 70 °C whereby the thiourea dissolves. The carboxylic acid chloride, prepared according to general procedure A, is also dissolved in toluene and this solution is slowly added to the warm solution of thiourea by a dropping funnel. After complete addition of the acid chloride, heat the reaction mixture for 1 h under reflux and leave it overnight at room temperature.

**General procedure D - Synthesis of piperazine monoamides** Piperazine (3-5 eq.) and PyBOP® (1.15 eq.) are dissolved in DMF and slowly added to a solution of the corresponding carboxylic acid in DMF. The reaction mixture is stirred overnight, then the solvent is removed under vacuum.
General procedure E Synthesis of thioureas with NaSCN

Dry NaSCN was suspended in acetone and cooled to 5 °C. An equimolar amount of benzoylchloride was dissolved in acetone and added dropwise. Subsequently, the mixture was stirred for 2 h at 5 °C. Equimolar amounts of the corresponding amine were dissolved in acetone, added dropwise at approx. 10 °C and the mixture stirred for 2 h at rt. After evaporation of the solvent, the residue was suspended in a small amount (approx. 4-8 eq.) of conc. HCl and heated to 90 °C for 1 h. After carefully neutralizing the mixture with conc. NH₃, the product was collected after precipitation via setting aside the mixture for 48 h at 8 °C or extracting the mixture with chloroform and subsequent flash chromatography of the combined organic layers.

General procedure F Synthesis of thioureas with trimethylsilylisothiocyanate

To a solution of the secondary (1 eq.) amine in dried THF trimethylsilylisothiocyanate (2 eq.) was added and stirred for 48 h. The solvent was removed under vacuum and a 2-propanol/water mixture (9:1) was added to the residue. The reaction mixture was boiled for a few seconds and the hot solution mixture was filtered. The flask and filter were washed twice with acetone. The filtrate was evaporated and the product was recrystallized from isopropanol/ diisopropyl ether (1:2).
**tert-Butylpiperazine-1-carboxylate (1a)** For the synthesis of 1a, piperazine (2000 mg, 23.26 mmol, 1.0 eq.) and triethylamine (4.837 ml, 34.90 mmol, 1.5 eq.) are dissolved in MeOH (20 ml), to which di-tert-butyl dicarbonate (2028 mg, 9.30 mmol, 0.4 eq.) is added slowly. The reaction mixture is then stirred at room temperature overnight. After completion of the reaction, the solvent is removed at the rotary evaporator and the crude product is purified by column chromatography (chloroform/MeOH 9:1). After purification the title compound is isolated as a colourless oil (1486 mg, 86 %). $^1$H-NMR (400 MHz, CDCl$_3$) δ 3.36 (t, CH$_2$-N(CO)-CH$_2$, $^2$J = 5.1 Hz, 4H), 2.78 (t, CH$_2$-NH-CH$_2$, $^2$J = 5.1 Hz, 4H), 1.74 (bs, CH$_2$-NH-CH$_2$, 1H), 1.44 (s, t-butyl, 9H) MS (ESI) m/z 187.36 [M+H$^+$]

**4-Carbamothioylpiperazine-1-carboxylate (1b)** Compound 1b is prepared by general procedure B, using tert-butylpiperazine-1-carboxylate (1a) (0.341 g, 1.83 mmol, 1.0 eq.) and thiocarbonyldiimidazole (0.376 g, 2.11 mmol, 1.15 eq.). Purification is carried out by MPLC (chloroform/MeOH gradient) and the title compound is isolated as a white solid (310 mg 69 %). $^1$H-NMR (400 MHz, CD$_3$OD) δ 3.83 (m, CH$_2$-N-CH$_2$, 4H), 3.48 (m, CH$_2$-NH-CH$_2$, 4H), 1.47 (s, t-butyl, 9H) MS (ESI) m/z 268.41 [M+Na$^+$]

**tert-Butyl 4-[8-nitro-4-oxo-6-(trifluoromethyl)-1,3-benzothiazin-2-yl]piperazine-1-carboxylate (1c)** 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (2.250 g, 8.4 mmol, 1.1 eq.) by general procedure A. 1c is synthesised by general procedure C, using 4-carbamothioylpiperazine-1-carboxylate (1b) (1.863 g, 7.6 mmol, 1.0 eq.) and adding DIPEA (3.878 ml, 22.8 mmol, 3.0 eq.). Purification is carried out by MPLC (Puriflash system, EtAc/heptane gradient). The title compound is isolated as yellow solid (3000 mg 86 %, HPLC purity 99.9 %, $t_R = 14.3$ min). $^1$H-NMR (400 MHz, CDCl$_3$) δ 9.09 (d, Ar-H, $^4$J = 2.2 Hz, 1H), 8.76 (d, Ar-H, $^4$J = 2.2 Hz, 1H), 4.01 (bs, CH$_2$-N-CH$_2$, 4H), 3.61 (bs, CH$_2$-N-CH$_2$, 4H), 1.49 (s, t-butyl, 9H) $^{13}$C-NMR (100 MHz, CDCl$_3$) δ
166.1, 162.5, 154.2, 143.8, 133.7, 133.3 (q, \(^3J_{CF} = 3.8\) Hz), 129.7 (q, \(^2J_{CF} = 35.9\) Hz), 126.6, 126.0 (q, \(^3J_{CF} = 3.8\) Hz), 122.1 (q, \(^4J_{CF} = 273.1\) Hz), 80.9, 46.1, 43.0, 28.5 HRMS \(m/z\) calc for C\(_{18}\)H\(_{20}\)F\(_3\)N\(_4\)O\(_5\)S [M+H\(^+\)], 461.1103; found, 461.1079, calc for C\(_{18}\)H\(_{19}\)F\(_3\)N\(_4\)O\(_5\)SNa [M+Na\(^+\)], 483.0921; found, 483.0896.

8-Nitro-2-(piperazin-1-yl)-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (1d) Compound (1c) (150 mg, 0.33 mmol, 1.0 eq.) is dissolved in a mixture of 0.5 ml DCM and 0.5 ml TFA and stirred for 1 h at room temperature. The solvent is removed under vacuum and the reaction mixture is co-evaporated three times each with toluene and chloroform. Compound 1d is isolated as a pale-yellow solid (106 mg 91 %) and used for the next reaction step without further purification. \(^1\)H-NMR (400 MHz, CD\(_3\)OD) \(\delta\) 8.88 (s, Ar-H, 1H), 8.84 (s, Ar-H, 1H), 4.29 (bs, CH\(_2\)-N-CH\(_2\), 4H), 3.49 (t, CH\(_2\)-NH\(_2\)-CH\(_2\), \(^3J = 5.3\) Hz, 4H) MS (ESI) \(m/z\) 361.63 [M+H\(^+\)]

8-Nitro-2-(4-octanoylpiperazin-1-yl)-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (1e) For the synthesis of 1e, caprylic acid (0.048 g, 0.33 mmol, 1.0 eq.) is dissolved in 10 ml DCM. DIPEA (0.166 ml, 0.98 mmol, 3.0 eq.), PyBOP\(^{\circ}\) (0.198 g, 0.38 mmol, 1.15 eq.) and 8-nitro-2-(piperazin-1-yl)-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (1d) (0.117 g, 0.33 mmol, 1.0 eq.) dissolved in 5 ml DCM are added to the reaction mixture. The mixture is then left overnight at room temperature and the solvent is removed in vacuo. Purification is carried out by MPLC (EtAc/Heptane gradient) and the title compound is isolated as a yellow solid (106 mg 59 %, HPLC purity 98.9 %, \(t_R = 15.2\) min). \(^1\)H-NMR (400 MHz, CDCl\(_3\)) \(\delta\) 9.08 (d, Ar-H, \(^4J = 2.0\) Hz, 1H), 8.76 (d, Ar-H, \(^4J = 2.0\) Hz, 1H), 4.03 (m, CH\(_2\)-N-CH\(_2\), 4H), 3.81 (bs, CH\(_2\)-N-CH\(_2\), 2H), 3.65 (bs, CH\(_2\)-N-CH\(_2\), 2H), 2.36 (t, CO-CH\(_2\), \(^3J = 7.6\) Hz, 2H), 1.64 (m, CH\(_2\), 2H), 1.30 (m, CH\(_2\), 8H), 0.88 (m, CH\(_3\)-CH\(_2\), 3H) \(^13\)C-NMR (100 MHz, CDCl\(_3\)) \(\delta\) 172.0, 166.2, 162.7, 143.9, 133.6, 133.5 (q, \(^3J_{CF} = 3.8\) Hz), 130.0 (q, \(^2J_{CF} = 35.1\) Hz), 126.6, 126.1 (q, \(^3J_{CF} = 3.8\) Hz), 122.2 (q, \(^4J_{CF} = 273.1\) Hz), 46.2, 46.0,
For the synthesis of 2a, stearic acid (0.062 g, 0.22 mmol, 1.0 eq.) is dissolved in 10 ml DCM. DIPEA (0.110 ml, 0.65 mmol, 3.0 eq.), PyBOP® (0.130 g, 0.25 mmol, 1.15 eq.) and 8-nitro-2-(piperazin-1-yl)-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (1d) (0.078 g, 0.22 mmol, 1.0 eq.), dissolved in 5 ml DCM, are added to the reaction mixture. The mixture is then left overnight at room temperature and the solvent is removed in vacuo. Purification is carried out by MPLC (EtAc/Heptane gradient) and the title compound is isolated as a yellow solid (71 mg 52%, HPLC purity 94.7%, t_R = 19.2 min). ^1H-NMR (500 MHz, CDCl_3) δ 9.10 (d, Ar-H, 4J = 2.0 Hz, 1H), 8.78 (d, Ar-H, 4J = 2.0 Hz, 1H), 4.05 (m, CH_2-N-CH_2, 4H), 3.82 (bs, CH_2-N-CH_2, 2H), 3.66 (bs, CH_2-N-CH_2, 2H), 2.37 (t, CO-C_6H_5, 3J = 7.6 Hz, 2H), 1.65 (m, CH_2, 2H), 1.29 (m, CH_2, 28H), 0.87 (m, CH_3-CH_2, 3H) ^13C-NMR (100 MHz, CDCl_3) δ 172.2, 166.3, 162.8, 143.9, 133.6, 133.5 (q, 3J_C,F = 3.8 Hz), 130.0 (q, 2J_C,F = 35.1 Hz), 126.5, 126.2 (q, 3J_C,F = 3.8 Hz), 122.2 (q, 1J_C,F = 273.1 Hz), 46.2, 46.0, 44.7, 40.8, 33.2, 31.9, 29.7, 29.6, 29.5, 29.4, 29.3, 25.1, 22.6, 14.1 HRMS m/z calc for C_{31}H_{46}F_3N_4O_4S [M+H]^+, 627.3187; found, 627.3177, calc for C_{31}H_{45}F_3N_4O_4Na [M+Na]^+, 649.3007; found, 649.2996.

1-(Piperazin-1-yl)-2-propylpentan-1-one (3a) The synthesis of 3a is carried out according to general procedure D. For this, sodium valproate (0.100 g, 0.60 mmol, 1.0 eq.) and PyBOP® (0.359 g, 0.69 mmol, 1.15 eq.) are dissolved in DMF (10 ml). Then a solution of piperazine (0.259 g, 3.01 mmol, 5.0 eq.) in DMF (5 ml) is added. After 16 h at RT, the solvent is removed in vacuo and the product is purified by column chromatography (chloroform/MeOH/ammonia lsg. 36% 90:10:1). The title compound is isolated as colourless oil (106 mg 84%). ^1H-NMR (400 MHz, CDCl_3) δ 3.58 (m, CH_2-N-CH_2, 2H), 3.47 (m, CH_2-N-CH_2, 2H), 2.78 (m, CH_2-N-CH_2, 4H), 2.37 (t, CO-C_6H_5, 3J = 7.6 Hz, 2H), 1.65 (m, CH_2, 2H), 1.29 (m, CH_2, 28H), 0.87 (m, CH_3-CH_2, 3H) ^13C-NMR (100 MHz, CDCl_3) δ 172.2, 166.3, 162.8, 143.9, 133.6, 133.5 (q, 3J_C,F = 3.8 Hz), 130.0 (q, 2J_C,F = 35.1 Hz), 126.5, 126.2 (q, 3J_C,F = 3.8 Hz), 122.2 (q, 1J_C,F = 273.1 Hz), 46.2, 46.0, 44.7, 40.8, 33.2, 31.9, 29.7, 29.6, 29.5, 29.4, 29.3, 25.1, 22.6, 14.1 HRMS m/z calc for C_{31}H_{46}F_3N_4O_4S [M+H]^+, 627.3187; found, 627.3177, calc for C_{31}H_{45}F_3N_4O_4Na [M+Na]^+, 649.3007; found, 649.2996.
4-(2-Propylpentanoyl)piperazin-1-thiourea (3b) 3b is prepared by general procedure B, using 1-(piperazin-1-yl)-2-propylpentan-1-one (3a) (0.106 g, 0.50 mmol, 1.0 eq.) and thiocarboxyldiimidazole (0.102 g, 0.58 mmol, 1.15 eq.). Purification is carried out by MPLC (heptane gradient) and the product is isolated as a white solid (98 mg 72%). $^1$H-NMR (400 MHz, CDCl$_3$) $\delta$ 6.23 (bs, NH$_2$, 2H), 4.03 (bs, CH$_2$-N-CH$_2$, 2H), 3.73 (bs, CH$_2$-N-CH$_2$, 4H), 3.66 (bs, CH$_2$-N-CH$_2$, 2H), 2.62 (m, CO-CH-(CH$_2$)$_2$, 1H), 1.58 (m, CH-CH$_2$, 2H), 1.38 (m, CH-CH$_2$, 2H), 1.22 (m, CH$_2$-CH$_3$, 4H), 0.86 (t, CH$_2$-CH$_3$, $^3$$J$ = 7.2 Hz, 6H) MS (APCI) m/z 272.2 [M+H$^+$].

8-Nitro-2-[4-(2-propylpentanoyl)piperazin-1-yl]-6-(trifluoromethyl)-4H-1,3-benzoazin-4-one (3c) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.055 g, 0.20 mmol, 1.1 eq.) by general procedure A. 3c is synthesised by general procedure C, using 4-(2-propylpentanoyl)piperazine-1-thiourea (3b) (0.050 g, 0.19 mmol, 1.0 eq.). Purification is carried out by MPLC (EtAc/heptane gradient) and the title compound is isolated as a yellow amorphous substance (84 mg 91 % HPLC purity 95.2 %, $t_R$ = 14.8 min). $^1$H-NMR (400 MHz, CDCl$_3$) $\delta$ 9.09 (d, Ar-H, $^4$$J$ = 2.2 Hz, 1H), 8.77 (d, Ar-H, $^4$$J$ = 2.2 Hz, 1H), 4.07 (bs, CH$_2$-N-CH$_2$, 2H), 3.98 (bs, CH$_2$-N-CH$_2$, 2H), 3.86 (bs, CH$_2$-N-CH$_2$, 2H), 3.74 (bs, CH$_2$-N-CH$_2$, 2H), 2.67 (m, CH$_2$-CH-CH$_2$, 1H), 1.65 (m, CH-CH$_2$, 2H), 1.43 (m, CH-CH$_2$, 2H), 1.27 (m, CH$_2$-CH$_3$, 4H), 0.89 (t, CH$_2$-CH$_3$, $^3$$J$ = 7.2 Hz, 6H) $^{13}$C-NMR (100 MHz, CDCl$_3$) $\delta$ 175.2, 166.2, 162.7, 143.9, 133.6, 133.5 (q, $^3$$J_{CF}$ = 3.8 Hz), 130.0 (q, $^2$$J_{CF}$ = 35.1 Hz), 126.6, 126.1 (q, $^3$$J_{CF}$ = 3.8 Hz), 122.3 (q, $^2$$J_{CF}$ = 273.1 Hz), 46.4, 46.2, 44.8, 41.0, 40.7, 35.2, 20.8, 14.2 HRMS m/z calc for C$_{31}$H$_{45}$F$_3$N$_4$O$_4$SNa [M+Na$^+$], 509.1445; found, 509.1450.

Ethyl-1-(piperazin-1-yl)hexan-1-one (4a) The synthesis of 4a is carried out according to general procedure D. For this, 2-ethylhexanoic acid (0.300 g, 2.08 mmol, 1.0 eq.) and PyBOP® (1.244

2.60 (m, CO-CH-(CH$_2$)$_2$, 1H), 2.00 (bs, NH, 1H), 1.58 (m, CH-CH$_2$, 2H), 1.32 (m, CH-CH$_2$, 2H), 1.20 (m, CH$_2$-CH$_3$, 4H), 0.82 (t, CH$_2$-CH$_3$, $^3$$J$ = 7.3 Hz, 6H) MS (APCI) m/z 213.2 [M+H$^+$].
g, 2.39 mmol, 1.15 eq.) are dissolved in DMF (20 ml). Then a solution of piperazine (0.537 g, 6.25 mmol, 3.0 eq.) in DMF (10 ml) is added. After 16 h at room temperature, the solvent is removed in vacuo and the product is purified by column chromatography (chloroform/MeOH/ammonia 36 % 90:10:1) and the title compound is isolated as colourless oil (284 mg 69 %). $^1$H-NMR (400 MHz, CDCl$_3$) δ 3.57 (t, CH$_2$-N-CH$_2$, $^3$$J$ = 4.9 Hz, 2H), 3.46 (t, CH$_2$-N-CH$_2$, $^3$$J$ = 4.9 Hz, 2H), 2.76 (m, CH$_2$-N-CH$_2$, 4H), 2.49 (m, CH$_2$-CH-CH$_2$, 1H), 2.02 (bs, NH, 1H), 1.57 (m, CH-CH$_2$, 2H), 1.37 (m, CH-CH$_2$, 2H), 1.16 (m, CH$_2$-CH$_2$, 6H) MS m/z (APCI) 213.2 [M+H$^+$].

4-(2-Ethylhexanoyl)piperazin-1-thiourea (4b) 4b is prepared according general procedure B, using Ethyl-1-(piperazin-1-yl)hexan-1-on (4a) (0.170 g, 0.80 mmol, 1.0 eq.) and thiocarbonyldiimidazole (0.164 g, 0.92 mmol, 1.15 eq.). Purification is carried out by MPLC (EtAc/heptane gradient) and the title compound is isolated as a white solid (132 mg 61 %). $^1$H-NMR (400 MHz, CD$_3$OD) δ 3.95 (bs, CH$_2$-N-CH$_2$, 2H), 3.81 (bs, CH$_2$-N-CH$_2$, 2H), 3.74 (m, CH$_2$-N-CH$_2$, 2H), 3.71 (m, CH$_2$-N-CH$_2$, 2H), 2.76 (m, CH$_2$-CH-CH$_2$, 1H), 1.60 (m, CH-CH$_2$, 2H), 1.48 (m, CH-CH$_2$, 2H), 1.27 (m, CH$_2$-CH$_3$, 4H), 0.90 (t, CH$_2$-CH$_3$, $^3$$J$ = 7.0 Hz, 3H), 0.88 (t, CH$_2$-CH$_3$, $^3$$J$ = 7.4 Hz, 3H) MS m/z (APCI) 272.3 [M+H$^+$].

2-[4-(2-Ethylhexanoyl)piperazin-1-yl]-8-nitro-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (4c) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.109 g, 0.41 mmol, 1.1 eq.) by general procedure A. 4c is synthesised by general procedure C, using 4-(2-ethylhexanoyl)piperazine-1-thiourea (4b) (0.100 g, 0.37 mmol, 1.0 eq.). Thet title compound is isolated by MPLC (EtAc/heptane gradient) (175 mg 97 %, HPLC purity 95.9 %, $t_R$ = 14.8 min). $^1$H-NMR (400 MHz, CDCl$_3$) δ 9.06 (d, Ar-H, $^4$$J$ = 1.8 Hz, 1H), 8.75 (d, Ar-H, $^4$$J$ = 1.8 Hz, 1H), 4.07 (bs, CH$_2$-N-CH$_2$, 2H), 3.97 (bs, CH$_2$-N-CH$_2$, 2H), 3.86 (bs, CH$_2$-N-CH$_2$, 2H), 3.73 (bs, CH$_2$-N-CH$_2$, 2H), 2.56 (m, CH$_2$-CH-CH$_2$, 1H), 1.65 (m, CH-CH$_2$,,
2H), 1.47 (m, CH-CH2, 2H), 1.23 (m, CH2-CH3, 4H), 0.87 (m, CH2-CH3, 6H) 13C-NMR (100 MHz, CDCl3) δ 175.1, 166.2, 162.7, 143.9, 133.6, 133.4 (q, JCF = 3.1 Hz), 130.0 (q, JCF = 35.1 Hz), 126.6, 126.1 (q, JCF = 3.1 Hz), 122.3 (q, JCF = 273.1 Hz), 46.4, 46.2, 44.8, 42.7, 41.0, 32.8, 29.8, 25.1, 22.8, 13.9, 12.0 HRMS m/z calc for C21H26F3N4O4S [M+H+] 487.1623; found, 487.1603, calc for C21H25F3N4O4SNa [M+Na+] 509.1442; found, 509.1420.

**tert-Butyl (S)-(4-methyl-1-oxo-1-(piperazin-1-yl)pentan-2-yl)carbamate (5a)**

The synthesis of 5a is carried out according to general procedure D. t-Boc-leucine (0.233 g, 1.01 mmol, 1.0 eq.) and PyBOP® (0.604 g, 1.16 mmol, 1.15 eq.) are dissolved in DMF (15 ml). Then a solution of piperazine (0.260 g, 3.02 mmol, 3.0 eq.) in DMF (5 ml) is added. After 16 h at room temperature, the solvent is removed in vacuo and the product is purified by column chromatography (chloroform/MeOH/ammonia 36:90:10:1). The title compound is a colourless oil (251 mg 84%). 1H-NMR (400 MHz, CDCl3) δ 5.31 (d, NH, J = 9.0 Hz, 1H), 4.60 (m, CO-CH-NH, 1H), 4.33 (bs, NH/H2O, 2H), 3.75 (m, CH2-N-CH2, 1H), 3.62 (m, CH2-N-CH2, 1H), 3.52 (m, CH2-N-CH2, 2H), 2.95 (m, CH2-N-CH2, 2H), 2.90 (m, CH2-N-CH2, 2H), 1.68 (m, CH3-CH-CH3, 1H), 1.43 (m, CH-CH2-CH, 2H), 1.40 (s, C-(CH3)3, 9H), 0.94 (d, CH3-CH-CH3, J = 6.7 Hz, 3H), 0.89 (d, CH3-CH-CH3, J = 6.7 Hz, 3H) MS m/z (APCI) 300.3 [M+H+].

**tert-Butyl (S)-(1-(4-carbamothioyloxy)piperazin-1-yl)-4-methyl-1-oxopentan-2-yl)carbamate (5b)**

Compound 5b is prepared by general procedure B, using tert-butyl N-[4-methyl-1-oxo-1-(piperazin-1-yl)pentan-2-yl]carbamate (5a) (0.202 g, 0.68 mmol, 1.0 eq.) and thiocarbonyldiimidazole (0.138 g, 0.78 mmol, 1.15 eq.). The product is purified by column chromatography (chloroform/MeOH 95:5) as a white solid. 1H-NMR (400 MHz, CDCl3) δ 6.25 (m, NH2, 2H), 5.25 (m, NH, 1H), 4.57 (m, CO-CH-NH, 1H), 4.07 (bs, CH2-N-CH2, 1H), 3.80 (bs, CH2-N-CH2, 5H), 3.55 (bs, CH2-N-CH2, 1H), 1.68 (m, CH3-CH-CH3, 1H), 1.45 (m, CH-CH2-CH, 2H), 1.30 (m, CH2-CH2-CH, 2H), 0.88 (d, CH3-CH3, J = 6.7 Hz, 3H).
1.39 (s, C-(CH₃)₃, 9H), 0.94 (d, CH₃-CH-CH₃, 3J = 6.5 Hz, 3H), 0.90 (d, CH₃-CH-CH₃, 3J = 6.6 Hz, 3H) MS m/z (ESI) 381.53 [M+Na⁺].

tert-Butyl (S)-(4-methyl-1-(4-(8-nitro-4-oxo-6-(trifluoromethyl)-4H-benzo[e][1,3]thiazin-2-yl)piperazin-1-yl)-1-oxopentan-2-yl)carbamate (5c) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.114 g, 0.42 mmol, 1.1 eq.) by general procedure A. AR160 is synthesised by general procedure C, using tert-butyl N-[1-(4-carbamothioylpiperazin-1-yl)-4-methyl-1-oxopentan-2-yl]carbamate (5b) (0.138 g, 0.39 mmol, 1.0 eq.). Purification of the title compound is done by column chromatography (chloroform/MeOH 98:2) (170 mg 76 %, HPLC purity 99.2 %, tᵣ = 14.8 min). ¹H-NMR (400 MHz, CDCl₃) δ 9.08 (d, Ar-H, 4J = 2.2 Hz, 1H), 8.76 (d, Ar-H, 4J = 2.2 Hz, 1H), 5.15 (d, N-H, 3J = 9.0 Hz, 1H), 4.64 (m, CO-CH-NH, 1H), 4.25 (bs, C-H₂-N-C-H₂, 2H), 4.04 (m, CH₂-N-CH₂, 1H), 3.94 (bs, CH₂-N-CH₂, 2H) 3.85 (m, CH₂-N-CH₂, 1H), 3.94 (bs, C-H₂-N-C-H₂, 2H), 3.85 (m, C-H₂-N-C-H₂, 1H), 3.59 (m, CH₂-N-CH₂, 2H), 1.70 (m, CH₃-CH-CH₃, 1H), 1.51 (m, CH₂-CH₂-CH, 1H), 1.42 (s, C-(CH₃)₃, 9H), 1.39 (m, CH-CH₂-CH, 1H), 0.97 (d, CH₃-CH-CH₃, 3J = 6.5 Hz, 3H), 0.93 (d, CH₃-CH-CH₃, 3J = 6.5 Hz, 3H) ¹³C-NMR (100 MHz, CDCl₃) δ 172.0, 166.2, 162.7, 155.6, 143.9, 133.6, 133.5 (q, 3Jₐₙ₈ₐ = 3.8 Hz), 130.0 (q, 3Jₐₙ₈₈ = 35.9 Hz), 126.6, 126.1 (q, 3Jₐₙ₈₈ = 3.8 Hz), 122.3 (q, 3Jₐₙ₈₈ = 273.1 Hz), 80.0, 48.4, 46.0, 44.8, 42.4, 41.5, 28.3, 24.7, 23.2, 22.0 HRMS m/z calc for C₂₄H₃₀F₃N₅O₆SNa [M+Na⁺], 596.1763; found, 596.1735.

2-(4-(L-Leucyl)piperazin-1-yl)-8-nitro-6-(trifluoromethyl)-4H-benzo[e][1,3]thiazin-4-one (5d) (47 mg, 0.08 mmol, 1.0 eq.) is dissolved in a mixture of 1 ml DCM and 1 ml TFA and left for 1 h at room temperature. The solvent is removed under vacuum and the reaction mixture is co-evaporated three times each with toluene and chloroform. The crude product is purified by column chromatography (chloroform/MeOH/ammonia 36 % 95:5:1) and the title compound is a yellow amorphous substance (31 mg 80 %, HPLC purity 97.4 %, tᵣ = 13.7 min). ¹H-NMR
(400 MHz, CDCl₃): δ = 9.09 (d, Ar-H, J = 2.0 Hz, 1H), 8.77 (d, Ar-H, J = 2.2 Hz, 1H), 4.04 (m, CH₂-N-CH₂, 5H), 3.76 (m, CH-NH₂, 1H), 3.73 (bs, CH₂-N-CH₂, 2H), 3.64 (bs, CH₂-N-CH₂, 1H), 1.86 (m, CH₃-CH-CH₃, 1H), 1.40 (m, CH-CH₂, 2H), 0.96 (dd, CH₃-CH-CH₃, J = 6.7 Hz, 6H) ¹³C-NMR (100 MHz, CDCl₃): δ = 174.6, 166.2, 162.8, 143.9, 133.6, 133.5 (q, J_C,F = 3.8 Hz), 129.1 (q, J_C,F = 35.1 Hz), 126.6, 126.2 (q, J_C,F = 3.8 Hz), 122.3 (q, J_C,F = 273.1 Hz), 49.7, 46.0, 44.8, 44.4, 41.4, 24.7, 23.5, 21.6 HRMS m/z calc for C₁₉H₂₃F₃N₅O₄ [M+H⁺], 474.1419; found, 474.1413.

4-(((tert-Butyloxy)carbonyl)amino)butanoic acid (6a) For the synthesis of 6a, γ-aminobutyric acid (1000 mg, 9.70 mmol, 1.0 eq.) and di-tert-butyl dicarbonate (2538 mg, 11.64 mmol, 1.2 eq.) are dissolved in THF (10 ml). To this solution a solution of NaHCO₃ (1076 mg, 12.80 mmol, 1.32 eq.) in water (10 ml) is added under ice cooling. The reaction mixture is then left at room temperature overnight. For work-up, the pH of the mixture is adjusted to 2 with 1 N HCl and the product is extracted with DCM (twice). The organic phases are collected and, after drying, concentrated over Na₂SO₄ in vacuo. The product - colourless oil - obtained is used without further purification (1453 mg 94%). ¹H-NMR (400 MHz, CDCl₃) δ 4.68 (bs, COO-H, 1H), 3.75 (m, NH, 1H), 3.18 (bs, NH-CH₂H), 2.39 (t, CO-CH₂, J = 7.4 Hz, 2H), 1.82 (m, CH₂-CH₂-CH₂, 2H), 1.52 (s, CH₃, 9H) MS m/z (ESI) 202.30 [M-H⁻].

tert-Butyl N-[4-oxo-4-(piperazin-1-yl)butyl]carbamate (6b) The synthesis of 6b is carried out according to general procedure D by dissolving 4-(((tert-butyloxy)carbonyl)amino)butanoic acid (6a) (1.453 g, 7.16 mmol, 1.0 eq.) and PyBOP® (4.282 g, 8.23 mmol, 1.15 eq.) in DMF (25 ml). Then a solution of piperazine (1.850 g, 21.47 mmol, 3.0 eq.) in DMF (10 ml) is added. After 16 h at room temperature the solvent is removed in vacuo and the product – a colourless oil - is purified by column chromatography (chloroform/MeOH/ammonia 36 % 90:10:1) (450 mg 23 %). ¹H-NMR (400 MHz, CDCl₃) δ 4.90 (bs, NH, 1H), 3.56 (m, CH₂-N-CH₂, 2H), 3.41 (m, CH₂-N-CH₂, 4H), 3.12 (q, NH-CH₂, J = 6.5 Hz, 2H), 2.83 (m, CH₂-N-CH₂, 4H), 2.77 (bs, NH, 1H), 2.32 (t,
CO-CH₂, 3J = 7.2 Hz, 2H), 1.78 (qi, CH₂-CH₂-CH₂, 3J = 7.0 Hz, 2H), 1.39 (s, CH₃, 9H) MS m/z (ESI) 294.18 [M+Na⁺].

tert-Butyl N-[4-{4-carbamothioylpiperazin-1-yl}-4-oxobutyl]carbamate (6c) Compound 6c is prepared by general procedure B, using tert-butyl N-[4-oxo-4-{piperazin-1-yl}butyl]carbamate (6b) (0.420 g, 1.55 mmol, 1.0 eq.) and thiocarbonyldiimidazole (0.318 g, 1.78 mmol, 1.15 eq.). The product - a white solid - is purified by column chromatography (chloroform/MeOH 9:1) (359 mg 70 %) ¹H-NMR (400 MHz, CDCl₃) δ 5.85 (bs, NH₂, 2H), 4.72 (bs, NH, 1H), 4.05 (bs, CH₂-N-CH₂, 2H), 3.75 (bs, CH₂-N-CH₂, 4H), 3.61 (bs, CH₂-N-CH₂, 2H), 3.18 (t, NH-CH₂, 3J = 6.6 Hz, 2H), 2.38 (t, CO-CH₂, 3J = 7.1 Hz, 2H), 1.85 (qi, CH₂-CH₂-CH₂, 3J = 6.8 Hz, 2H), 1.43, (s, CH₃, 9H) MS m/z (ESI) 353.66 [M+Na⁺].

tert-Butyl N-[4-{4-[8-nitro-4-oxo-6-(trifluoromethyl)-4H-1,3-benzothiazin-2-yl]piperazin-1-yl}-4-oxobutyl]carbamate (6d) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.436 g, 1.62 mmol, 1.1 eq.) by general procedure A. 6d is synthesised by general procedure C, using tert-butyl N-[4-{4-carbamothioylpiperazin-1-yl}-4-oxobutyl]carbamate (6c) (0.138 g, 0.39 mmol, 1.0 eq.). Purification is carried out by MPLC (chloroform/MeOH gradient) and the title compound is isolated as a yellow solid (195 mg 24 %, HPLC purity 95.4 %, tᵣ = 13.7 min). ¹H-NMR (400 MHz, CDCl₃) δ 9.08 (d, Ar-H, 4J = 2.2 Hz, 1H), 8.76 (d, Ar-H, 4J = 2.2 Hz, 1H), 4.73 (bs, NH, 1H), 4.04 (m, CH₂-N-CH₂, 4H), 3.82 (bs, CH₂-N-CH₂, 2H), 3.65 (bs, CH₂-N-CH₂, 2H), 3.18 (q, NH-CH₂, 3J = 6.5 Hz, 2H), 2.41 (t, CO-CH₂, 3J = 7.1 Hz, 2H), 1.83 (qi, CH₂-CH₂-CH₂-CH₂, 3J = 6.8 Hz, 2H), 1.42, (s, CH₃, 9H) ¹³C-NMR (100 MHz, CDCl₃) δ 171.3, 166.1, 162.7, 156.1, 143.9, 133.6, 133.6 (q, 3J_C,F = 3.1 Hz), 129.9 (q, 2J_C,F = 35.9 Hz), 126.5, 126.1 (q, 3J_C,F = 3.8 Hz), 122.3 (q, 1J_C,F = 273.1 Hz), 79.1, 46.1, 45.9, 44.6, 40.9, 40.1, 30.2, 28.4, 25.3 HRMS m/z calc for C₂₂H₂₆F₃N₅O₆SNa [M+Na⁺], 568.1449; found, 568.1442.
(S)-N-(4-(Methylthio)-1-oxo-1-(piperazin-1-yl)butan-2-yl)acetamide (7a) The synthesis of 7a is carried out according to general procedure D. For this, N-acetylcysteine (0.244 g, 1.50 mmol, 1.0 eq.) and PyBOP® (0.897 g, 1.73 mmol, 1.15 eq.) are dissolved in DMF (15 ml). Then a solution of piperazine (0.388 g, 4.50 mmol, 3.0 eq.) in DMF (5 ml) is added. After 16 h at room temperature, the solvent is removed in vacuo and the product is purified by column chromatography (chloroform/MeOH/ammonia 36% 90:10:1). The title compound is isolated as a colourless oil (159 mg 41%). 1H-NMR (400 MHz, CD3OD) δ 6.67 (d, NH, J = 8.0 Hz, 1H), 5.05 (m, CO-C-H-N, 1H), 3.56 (m, CH2-N-CH2, 1H), 2.85 (m, CH2-N-CH2, 1H), 2.47 (m, CH2-CH2-S, 2H), 2.06 (s, -CH3, 3H), 1.98 (s, -CH3, 3H), 1.95 (m, CH-CH2-CH2, 1H), 1.79 (m, CH-CH2-CH2, 1H) MS m/z (ESI) 260.64 [M+H]+, 283.15 [M+Na+].

(S)-N-(1-(4-Carbamothioyl)piperazin-1-yl)-4-(methylthio)-1-oxobutan-2-yl)acetamide (7b) Compound 7b is prepared by general procedure B, using N-[4-(methylsulfanyl)-1-oxo-1-(piperazin-1-yl)butan-2-yl]acetamide (7a) (0.159 g, 0.61 mmol, 1.0 eq.) and thiocarbonyldiimidazole (0.126 g, 0.71 mmol, 1.15 eq.). The purification is carried out by recrystallisation from DCM/hexane 1:1 and the title compound is a white solid (116 mg 60%). 1H-NMR (400 MHz, CD3OD) δ 4.98 (m, CO-C-H-N, 1H), 4.03 (m, CH2-N-CH2, 1H), 3.94 (m, CH2-N-CH2, 1H), 3.86 (m, CH2-N-CH2, 1H), 3.75 (m, CH2-N-CH2, 1H), 3.55 (m, CH2-N-CH2, 1H), 2.53 (m, CH2-CH2-S, 2H), 2.10 (s, -CH3, 3H), 1.99 (m, CH-CH2-CH2, 1H), 1.97 (s, -CH3, 3H), 1.90 (m, CH-CH2-CH2, 1H) MS m/z (ESI) 341.50 [M+Na+].

(S)-N-[4-(Methylthio)-1-(4-(8-nitro-4-oxo-6-(trifluoromethyl)-4H-benzo[e][1,3]thiazin-2-yl)piperazin-1-yl)-1-oxobutan-2-yl]acetamide (7c) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.093 g, 0.35 mmol, 1.1 eq.) by general procedure A. 7c is synthesised by general procedure C, using N-[1-(4-carbamothioyl)piperazin-1-yl)-4-(methylsulfanyl)-1-oxobutan-2-yl]acetamide (7b) (0.100 g,
0.31 mmol, 1.0 eq.). Purification is carried out by MPLC (EtAc/MeOH gradient) and the title compound is isolated as a yellow amorphous substance (73 mg 44 %, HPLC purity 100.0 %, \( t_R = 12.3 \text{ min} \)). \( ^1H \)-NMR (400 MHz, CDCl\(_3\)) \( \delta \) 9.04 (s, Ar-H, 1H), 8.74 (s, Ar-H, 1H), 6.66 (d, NH, \( ^3J = 7.2 \text{ Hz}, 1\text{H} \)), 5.10 (m, CO-CH-NH, 1H), 4.18 (bs, CH\(_2\)-N-CH\(_2\), 2H), 3.98 (m, CH\(_2\)-N-CH\(_2\), 4H), 3.76 (m, CH\(_2\)-N-CH\(_2\), 1H), 3.61 (m, CH\(_2\)-N-CH\(_2\), 2H), 2.51 (m, CH\(_2\)-S, 2H), 2.07 (s, -CH\(_3\), 3H), 1.99 (s, -CH\(_3\), 3H), 1.98 (m, CH-CH-CH\(_2\), 1H), 1.87 (m, CH\(_2\)-S, 1H) \( ^{13}C \)-NMR (100 MHz, CDCl\(_3\)) \( \delta \) 175.1, 166.2, 162.7, 143.9, 133.6, 133.4 (q, \( ^3J_{C,F} = 3.1 \text{ Hz} \)), 130.0 (q, \( ^2J_{C,F} = 35.1 \text{ Hz} \)), 126.6, 126.1 (q, \( ^3J_{C,F} = 3.1 \text{ Hz} \)), 122.3 (q, \( ^1J_{C,F} = 273.1 \text{ Hz} \), 46.4, 46.2, 44.8, 42.7, 41.0, 32.8, 29.8, 25.8, 22.8, 13.9, 12.0 HRMS m/z calc for C\(_{20}\)H\(_{23}\)F\(_3\)N\(_5\)O\(_5\)S\(_2\) [M+H\(^+\)], 534.1089; found, 534.1062, calc for C\(_{20}\)H\(_{22}\)F\(_3\)N\(_5\)O\(_5\)S\(_2\)Na [M+Na\(^+\)], 556.09118; found, 556.0881.

2-[4-rac-(2-Methylpropyl)phenyl]-1-(piperazin-1-yl)propan-1-one (8a) The synthesis of 8a is carried out according to general procedure D by dissolving 2-[4-(2-methylpropyl)phenyl]propanoic acid (0.500 g, 2.42 mmol, 1.0 eq.) and PyBOP\(^\circ\) (1.450 g, 2.79 mmol, 1.15 eq.) in DMF (10 ml). Then a solution of piperazine (0.626 g, 7.27 mmol, 3.0 eq.) in DMF (10 ml) is added. After 16 h at room temperature the solvent is removed in vacuo and the product is purified by column chromatography (chloroform/MeOH/ammonia 36 % 90:10:1) as a colourless oil (534 mg 80 %). \( ^1H \)-NMR (400 MHz, CDCl\(_3\)) \( \delta \) 7.06 (m, Ar-H, 4H), 6.88 (bs, NH, 1H), 3.94 (bs, CH\(_2\)-N-CH\(_2\), 1H) 3.76 (q, CO-CH, \( ^3J = 6.8 \text{ Hz}, 1\text{H} \)), 3.47 (m, CH\(_2\)-N-CH\(_2\), 3H), 3.04 (bs, CH\(_2\)-N-CH\(_2\), 1H), 2.85 (m, CH\(_2\)-N-CH\(_2\), 2H), 2.40 (d, CH\(_2\), \( ^3J = 7.2 \text{ Hz}, 2\text{H} \)), 2.34 (bs, CH\(_2\)-N-CH\(_2\), 1H), 1.80 (n, CH\(_2\)-CH-(CH\(_3\))\(_2\), \( ^3J = 6.8 \text{ Hz}, 1\text{H} \)), 1.38 (d, CH\(_3\)-CH, \( ^3J = 6.8 \text{ Hz}, 3\text{H} \)), 0.85 (d, CH\(_3\)-CH\(_3\), \( ^3J = 6.7 \text{ Hz}, 6\text{H} \)) MS m/z (APCI) 275.2 [M+H\(^+\)].

4-[2-[4-rac-(2-Methylpropyl)phenyl]propanoyl]piperazin-1-thiourea (8b) Compound 8b is prepared by general procedure B, using 2-[4-(2-methylpropyl)phenyl]-1-(piperazin-1-yl)propan-1-one (8a) (0.534 g, 1.94 mmol, 1.0 eq.) and thiocarbonyldiimidazole (0.397 g, 2.23
mmol, 1.15 eq.). The product is recrystallised from isopropanol/diisopropyl ether (1:1) and the title compound is a white solid (407 mg 63 %). $^1$H-NMR (400 MHz, CD$_3$OD) δ 7.17 (d, Ar-H, $^3$J = 8.2 Hz, 2H), 7.12 (d, Ar-H, $^3$J = 8.2 Hz, 2H), 4.04 (q, CO-CH, $^3$J = 6.8 Hz, 1H), 3.85 (m, CH$_2$-N-CH$_2$, 3H), 3.52 (m, CH$_2$-N-CH$_2$, 2H), 3.16 (m, CH$_2$-N-CH$_2$, 1H), 2.45 (d, CH$_3$, $^3$J = 7.2 Hz, 2H), 1.83 (n, CH$_2$-CH-(CH$_3$)$_2$, $^3$J = 6.8 Hz, 1H), 1.38 (d, CH$_3$-CH, $^3$J = 6.8 Hz, 3H), 0.88 (d, CH$_3$-CH-CH$_3$, $^3$J = 6.7 Hz, 6H) MS m/z (ESI) 356.86 [M+H$^+$].

2-(4-[(2R)-[4-(2-Methylpropyl)phenyl]propanoyl]piperazin-1-yl]-8-nitro-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (8c) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.133 g, 0.50 mmol, 1.1 eq.) by general procedure A. Compound 8c is synthesised by general procedure C, using 4-[2-[4-(2-methylpropyl)phenyl]propanoyl]piperazine-1-thiourea (8b) (0.138 g, 0.39 mmol, 1.0 eq.). Purification is carried out by MPLC (EtAc/heptane gradient) and the title compound is isolated as a yellow solid (207 mg 84 %, HPLC purity 98.9 %, $t_R$ = 15.9 min). $^1$H-NMR (400 MHz, CDCl$_3$) δ 9.07 (d, Ar-H, $^4$J = 2.0 Hz, 1H), 8.76 (d, Ar-H, $^4$J = 2.0 Hz, 1H), 7.14 (d, Ar-H, $^3$J = 8.4 Hz, 2H), 7.11 (d, Ar-H, $^3$J = 8.4 Hz, 2H), 4.15 (bs, CH$_2$-N-CH$_2$, 3H), 3.84 (q, CO-CH, $^3$J = 6.8 Hz, 1H), 3.60 (m, CH$_2$-N-CH$_2$, 2H), 3.48 (m, CH$_2$-N-CH$_2$, 2H), 3.06 (bs, CH$_2$-N-CH$_2$, 1H), 2.44 (d, CH$_3$, $^3$J = 7.2 Hz, 2H), 1.84 (n, CH$_2$-CH-(CH$_3$)$_2$, $^3$J = 6.8 Hz, 1H), 1.47 (d, CH$_3$-CH, $^3$J = 6.8 Hz, 3H), 0.88 (dd, CH$_3$-CH-CH$_3$, $^3$J = 6.5 Hz, 6H) $^{13}$C-NMR (100 MHz, CDCl$_3$) δ 172.5, 166.1, 162.5, 143.9, 140.8, 138.5, 133.6, 133.4 (q, $^3$J$_{CF}$ = 3.8 Hz), 130.0, 129.9 (q, $^3$J$_{CF}$ = 35.1 Hz), 126.8, 126.6, 126.1 (q, $^3$J$_{CF}$ = 3.8 Hz), 122.3 (q, $^3$J$_{CF}$ = 273.1 Hz), 46.1, 45.6, 44.9, 44.8, 43.4, 41.4, 30.1, 22.3, 20.5 HRMS m/z calc for C$_{26}$H$_{28}$F$_3$N$_4$O$_5$ [M+H$^+$], 549.1779; found, 549.1764, calc for C$_{26}$H$_{26}$F$_3$N$_4$O$_5$Na [M+Na$^+$], 571.1599; found, 571.1583.

2-[4-[(2S)-2-[4-(2-Methylpropyl)phenyl]propanoyl]piperazin-1-yl]-8-nitro-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (9a) For the synthesis of 9a, (2S)-2-[4-(2-
methyl[propyl]phenyl]propanoic acid (0.068 g, 0.33 mmol, 1.0 eq.) is dissolved in 10 ml DCM. DIPEA (0.166 ml, 0.98 mmol, 3.0 eq.), PyBOP® (0.198 g, 0.38 mmol, 1.15 eq.) and 8-nitro-2-(piperazin-1-yl)-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (1d) (0.117 g, 0.33 mmol, 1.0 eq.) dissolved in 5 ml DCM are added to the reaction mixture. The mixture is then left overnight at room temperature and the solvent is removed in vacuo. Purification is carried out by MPLC (EtAc/Heptane gradient). The title compound is a yellow solid (164 mg 91 %, HPLC purity 97.4 %, t\textsubscript{R} = 15.9 min). \textsuperscript{1}H-NMR (400 MHz, CDCl\textsubscript{3}) \(\delta\) 9.06 (d, Ar-H, \textsubscript{4}J = 2.0 Hz, 1H), 8.75 (d, Ar-H, \textsubscript{4}J = 2.0 Hz, 1H), 7.14 (d, Ar-H, \textsubscript{3}J = 8.2 Hz, 2H), 7.11 (d, Ar-H, \textsubscript{3}J = 8.2 Hz, 2H), 4.15 (m, C\textsubscript{H}\textsubscript{2}-N-C\textsubscript{H}\textsubscript{2}, 3H), 3.84 (q, CO-C\textsubscript{H}, \textsubscript{3}J = 6.8 Hz, 1H), 3.61 (m, C\textsubscript{H}\textsubscript{2}-N-C\textsubscript{H}\textsubscript{2}, 2H), 3.48 (m, C\textsubscript{H}\textsubscript{2}-N-C\textsubscript{H}\textsubscript{2}, 2H), 3.06 (bs, C\textsubscript{H}\textsubscript{2}-N-C\textsubscript{H}\textsubscript{2}, 1H), 2.43 (d, C\textsubscript{H}\textsubscript{2}, \textsubscript{3}J = 7.2 Hz, 2H), 1.84 (n, CH\textsubscript{2}-C\textsubscript{H}-(CH\textsubscript{3})\textsubscript{2}, \textsubscript{3}J = 6.8 Hz, 1H), 1.46 (d, C\textsubscript{H}\textsubscript{3}-CH, \textsubscript{3}J = 6.8 Hz, 3H), 0.87 (d, C\textsubscript{H}\textsubscript{3}-CH-C\textsubscript{H}\textsubscript{3}-C\textsubscript{H}\textsubscript{3}, \textsubscript{3}J = 6.7 Hz, 6H) \textsuperscript{13}C-NMR (100 MHz, CDCl\textsubscript{3}) \(\delta\) 172.5, 166.1, 162.5, 143.9, 140.8, 138.5, 133.6, 133.4 (q, \textsubscript{3}J\textsubscript{C,F} = 3.8 Hz), 130.0, 129.9 (q, \textsubscript{2}J\textsubscript{C,F} = 35.1 Hz), 126.8, 126.6, 126.1 (q, \textsubscript{3}J\textsubscript{C,F} = 3.8 Hz), 122.2 (q, \textsubscript{1}J\textsubscript{C,F} = 273.1 Hz), 46.0, 45.6, 44.9, 44.8, 43.4, 41.4, 30.1, 22.3, 20.5 HRMS m/z calc for C\textsubscript{26}H\textsubscript{28}F\textsubscript{3}N\textsubscript{4}O\textsubscript{4}S [M+H\textsuperscript{+}], 549.1779; found, 549.1783, calc for C\textsubscript{26}H\textsubscript{27}F\textsubscript{3}N\textsubscript{4}O\textsubscript{4}SNa [M+Na\textsuperscript{+}], 571.1599; found, 571.1600.

\textsuperscript{1}-(4-Methylbenzensulfonyl)piperazine (10a) p-Toluenesulfonic acid chloride (0.500 g, 2.62 mmol, 1.0 eq.) is taken up in DMF (10 ml), added to a solution of piperazine (1.552 g, 13.05 mmol, 5.0 eq.) in DMF (20 ml) and stirred overnight (inert gas). The solvent is removed under vacuum and the crude product is purified by column chromatography (chloroform/Methanol/ammonia 36 % 90:10:1). The title compound is isolated as a colourless oil (458 mg 73 %). \textsuperscript{1}H-NMR (400 MHz, CDCl\textsubscript{3}) \(\delta\) 7.55 (dd, Ar-H, \textsubscript{3}J = 8.4 Hz, \textsubscript{4}J = 2.0 Hz 2H), 7.25 (d, Ar-H, \textsubscript{3}J = 8.4 Hz, 2H), 2.87 (m, CH\textsubscript{2}-N-CH\textsubscript{2}, 8H), 2.36 (s, CH\textsubscript{3}, 3H) MS m/z (APCI) 241.1 [M+H\textsuperscript{+}].

\textsuperscript{4}-(4-methylbenzensulfonyl)piperazin-1-thiourea (10b) Compound 10b is prepared by general procedure B, using \textsuperscript{1}-(4-methylbenzenesulfonyl)piperazine (10a) (0.400 g, 1.66 mmol, 1.0 eq.)
and thiocarbonyldiimidazole (0.341 g, 1.19 mmol, 1.15 eq.). The product is recrystallised as a white solid from isopropanol/diisopropyl ether (1:1) (172 mg 35%). $^1$H-NMR (400 MHz, DMSO-D$_6$) δ 7.63 (d, Ar-H, $^3$J = 8.2 Hz, 2H) 7.49 (bs, NH$_2$, 2H) 7.46 (d, Ar-H, $^3$J = 8.0 Hz, 2H), 3.84 (bs, CH$_2$-N-CH$_2$, 4H), 2.41 (s, CH$_3$, 3H) MS m/z (ESI) 322.70 [M+Na$^+$].

2-[4-(4-Methylbenzenesulfonyl)piperazin-1-yl]-8-nitro-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (10c) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.149 g, 0.55 mmol, 1.1 eq.) by general procedure A. 10c is synthesised by general method 3, using 4-{24-(4-methylbenzenesulfonyl)piperazine-1-thiourea (10b) (0.150 g, 0.50 mmol, 1.0 eq.). Purification is carried out by MPLC (EtAc/Heptane gradient). The title compound is isolated as a yellow solid (213 mg 86%, HPLC purity 98.9%, $t_R = 14.0$ min) $^1$H-NMR (400 MHz, CDCl$_3$) δ 9.04 (d, Ar-H, $^4$J = 2.2 Hz, 1H), 8.74 (d, Ar-H, $^4$J = 2.2 Hz, 1H), 7.64 (d, Ar-H, $^3$J = 8.4 Hz, 2H), 7.34 (d, Ar-H, $^3$J = 8.4 Hz, 2H), 4.11 (bs, CH$_2$-N-CH$_2$, 4H), 3.17 (m, CH$_2$-N-CH$_2$, 4H), 2.42 (s, CH$_3$, 3H) $^{13}$C-NMR (100 MHz, DMSO-d$_6$) δ 165.7, 162.5, 144.8, 144.4, 134.6, 132.3, 131.8 (q, $^3$J$_{C,F}$ = 3.8 Hz), 130.4, 128.0, 127.9 (q, $^2$J$_{C,F}$ = 34.3 Hz), 126.7 (q, $^3$J$_{C,F}$ = 3.8 Hz), 126.5, 123.0 (q, $^2$J$_{C,F}$ = 273.7 Hz), 45.8, 45.6, 21.4 HRMS m/z calc for C$_{20}$H$_{18}$F$_3$N$_4$O$_5$S$_2$ [M+H$^+$], 515.0667; found, 515.0665, calc for C$_{20}$H$_{17}$F$_3$N$_4$O$_5$S$_2$Na [M+Na$^+$], 537.0486; found, 537.0483.

1-(Heptane-1-sulfonyl)piperazine (11a) Sodium-1-heptanesulfonate (1.50 g, 6.81 mmol, 1.0 eq.) is mixed with POCl$_3$ (4.66 ml, 51.19 mol, 7.5 eq.) in a round bottom flask and heated to 100 °C for 1 h with vigorous stirring. Then cool the reaction mixture to 0 °C, add 10 ml toluene and 25 ml water. The organic phase is separated, washed twice with water and concentrated in vacuo after drying over NaSO$_4$. The acid chloride is taken up without further purification in DMF (15 ml), added to a solution of piperazine (4.05 g, 34.05 mmol, 5.0 eq.) in DMF (35 ml) and stirred overnight (inert gas). The solvent is removed under vacuum and the crude product
is worked up by column chromatography (chloroform/Methanol/ammonia 36% 90:10:1). The
product is isolated as a colourless oil (880 mg 65%). $^1$H-NMR (400 MHz, CDCl$_3$) $\delta$ 3.23 (m, CH$_2$-
N-CH$_2$, 4H), 2.91 (m, CH$_2$-N-CH$_2$, 4H), 2.86 (m, SO$_2$-CH$_2$, 2H), 2.34 (bs, NH, 1H), 1.78 (m, CH$_2$,
2H), 1.38 (m, CH$_2$, 2H), 1.26 (m, CH$_2$, 6H), 0.86 (m, CH$_3$, 3H) MS m/z (APCI) 249.2 [M+H$^+$].

4-(heptane-1-sulfonyl)piperazin-1-thiourea (11b) Compound 11b is prepared by general
procedure B, using 1-(heptane-1-sulfonyl)piperazine (11a) (0.300 g, 1.21 mmol, 1.0 eq.) and
thiocarbonyldiimidazole (0.248 g, 1.39 mmol, 1.15 eq.). Purification is carried out by MPLC
(chloroform /MeOH gradient). The product is a white compound (311 mg 84%). $^1$H-NMR
(400 MHz, CDCl$_3$) $\delta$ 5.92 (bs, NH, 1H), 3.93 (m, CH$_2$-N-CH$_2$, 4H), 3.37 (t, CH$_2$-N-CH$_2$,
4J = 5.1 Hz, 4H), 2.92 (m, SO$_2$-CH$_2$, 2H), 1.79 (m, CH$_2$, 2H), 1.41 (m, CH$_2$, 2H), 1.29 (m, CH$_2$, 6H), 0.88 (m,
CH$_3$, 3H) MS m/z (ESI) 330.85 [M+Na$^+$].

2-[4-(Heptane-1-sulfonyl)piperazine-1-yl]-8-nitro-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-
one (11c) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-
3-nitro-5-(trifluoromethyl)benzoic acid (0.145 g, 0.54 mmol, 1.1 eq.) by general procedure A.
Compound 11c is synthesised by general procedure C, using 4-(heptane-1-sulfonyl)piperazine-
1-thiourea (11b) (0.150 g, 0.49 mmol, 1.0 eq.). Purification is carried out by MPLC
(EtAc/heptane gradient). The title compound is slightly yellow (234 mg 91%, HPLC purity
99.5%, $t_R = 15.1$ min). $^1$H-NMR (400 MHz, CDCl$_3$) $\delta$ 9.09 (d, Ar-H, 4J = 2.2 Hz, 1H), 8.78 (d, Ar-
H, 4J = 2.2 Hz, 1H), 4.12 (bs, CH$_2$-N-CH$_2$, 4H), 3.47 (m, CH$_2$-N-CH$_2$, 4H), 2.95 (m, SO$_2$-CH$_2$, 2H),
1.81 (m, CH$_2$, 2H), 1.42 (m, CH$_2$, 2H), 1.29 (m, CH$_2$, 6H), 0.88 (m, CH$_3$, 3H) $^{13}$C-NMR (100 MHz,
CDCl$_3$) $\delta$ 166.3, 162.7, 143.9, 133.5 (q, $^3$J$_{CF} = 3.8$ Hz), 133.4, 130.1 (q, $^2$J$_{CF} = 35.9$ Hz), 126.6,
126.2 (q, $^3$J$_{CF} = 3.8$ Hz), 122.3 (q, $^1$J$_{CF} = 273.1$ Hz), 50.4, 46.5, 45.4, 31.4, 28.7, 28.3, 23.1, 22.5,
14.0 HRMS m/z calc for C$_{20}$H$_{26}$F$_3$NaO$_5$S$_2$ [M+H$^+$], 523.1292; found, 523.1279, calc for
C$_{20}$H$_{25}$F$_3$NaO$_5$SNa [M+Na$^+$], 545.1112; found, 545.1098.
Pyrrolidine-1-thiourea (12a) Compound 12a is prepared by general procedure B, using pyrrolidine (0.450 g, 6.33 mmol, 1.0 eq.) and thiocarbonyldiimidazole (1.300 g, 7.28 mmol, 1.15 eq.). The crude product is worked up by column chromatography (chloroform/MeOH 98:2). The title compound is a white solid (187 mg 21%). $^1$H-NMR (400 MHz, CDCl$_3$) δ 5.71 (bs, NH$_2$, 2H), 3.82 (bs, N-CH$_2$, 2H), 3.38 (bs, N-CH$_2$, 2H), 2.07 (bs, CH$_2$-CH$_2$, 2H), 1.95 (bs, CH$_2$-CH$_2$, 2H) MS m/z (APCI) 131.1 [M+H$^+$].

8-Nitro-2-(pyrrolidine-1-yl)-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (12b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.210 g, 0.77 mmol, 1.1 eq.) by general procedure A. Compound 12b is synthesised by general procedure C, using pyrrolidine-1-thiourea (12a) (0.100 g, 0.70 mmol, 1.0 eq.). The crude product is worked up by column chromatography (LM chloroform) and washed with diisopropyl ether. The title compound is isolated a yellow solid (85 mg 34%, HPLC purity 99.1 %, $t_R$ = 13.5 min). $^1$H-NMR (400 MHz, CDCl$_3$) δ 9.17 (d, Ar-H, $^4$J = 2.2 Hz, 1H), 8.77 (d, Ar-H, $^4$J = 2.2 Hz, 1H), 3.93 (t, CH$_2$-N-CH$_2$, $^3$J = 6.9 Hz, 2H), 3.70 (t, CH$_2$-N-CH$_2$, $^3$J = 6.9 Hz, 2H), 2.19 (qi, CH$_2$-CH$_2$, $^3$J = 6.9 Hz, 2H), 2.06 (bs, CH$_2$-CH$_2$, $^3$J = 6.9 Hz, 2H) $^{13}$C-NMR (100 MHz, CDCl$_3$) δ 165.9, 160.3, 143.5, 134.6, 133.7 (q, $^1$J$_{C,F}$ = 3.8 Hz), 129.5 (q, $^2$J$_{C,F}$ = 35.9 Hz), 126.9, 125.8 (q, $^3$J$_{C,F}$ = 3.8 Hz), 122.4 (q, $^1$J$_{C,F}$ = 273.1 Hz), 50.4, 47.4, 25.5, 24.1 HRMS m/z calc for C$_{13}$H$_{11}$F$_3$N$_2$O$_5$S [M+H$^+$], 346.0469; found, 346.0461.

Piperidine-1-thiourea (13a)

Synthesis by procedure B Compound 13a is prepared by general procedure B, using piperidine (1.000 g, 11.74 mmol, 1 eq.) and thiocarbonyldiimidazole (2.406 g, 13.50 mmol, 1.15 eq.). The crude product is purified by column chromatography (chloroform/MeOH 98:2) and the title compound is isolated as a white solid (723 mg 43%).
Synthesis by procedure E Synthesis according to general procedure E, starting from piperidine (4.95 ml, 50 mmol). Work-up after neutralization: the mixture was kept at 8 °C for 48 h, the brown oil which settled on the bottom of the flask was separated and purified by flash chromatography twice (eluent chloroform). The fractions containing the product were combined, the solvent evaporated and the remaining crude product treated with a few ml of toluene. A white precipitate formed, which was filtered off and dried (611 mg 9 %).

Synthesis by procedure F Synthesis according to general procedure F, starting from piperidine (49 mg, 0.57 mmol, 1 eq.). Compound 13a was isolated as a slightly brownish solid (30 mg, 36 %)

1H-NMR (400 MHz, CD3OD) δ 3.78 (bs, CH2-N-CH2, 4H), 1.67 (m, CH2, 2H), 1.61 (m, CH2, 4H) MS m/z (ESI) 145.09 [M+H+].

8-Nitro-2-(piperidine-1-yl)-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (13b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (1.116 g, 4.14 mmol, 1.0 eq.) by general procedure A. Compound 13b is synthesised by general procedure C, using piperidine-1-thiourea (13a) (0.597 g, 4.14 mmol, 1.0 eq.). The crude product is processed by column chromatography (chloroform) and the titel compound appears as a yellow solid (1105 mg 74 %, HPLC purity 96.2 %, tR = 14.1 min). 1H-NMR (400 MHz, CDCl3) δ 9.09 (d, Ar-H, 4J = 2.5 Hz, 1H), 8.74 (d, Ar-H, 4J = 2.5 Hz, 1H), 3.97 (bs, N-C-H2, 4H), 1.77 (m, CH2-CH2-CH2, 6H) 13C NMR (126 MHz, CDCl3) δ 166.6, 161.5, 143.9, 134.3, 133.3 (q, 3JCF = 3.5 Hz), 129.5 (d, 2JCF = 35.3 Hz), 126.7, 125.9 (q, 3JCF = 3.6 Hz), 122.4 (q, 3JCF = 272.5 Hz), 47.9, 26.0, 24.3 MS m/z (APCI) 360.2 [M+H+].

2-(2,6-Dimethylpiperidin-1-yl)-8-nitro-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (14a) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.500 g, 1.86 mmol, 1.0 eq.) by general procedure A. The 2-
chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is dissolved in acetone (5 ml) and added to a suspension of KSCN (0.181 g, 1.86 mmol, 1.0 eq.) in acetone (10 ml) at 5 °C under inert gas. The reaction mixture is stirred for 2 h at 5-10 °C. Dimethylpiperidine (0.210 g, 1.86 mmol, 1 eq.) is then dissolved in acetone (5 ml) and added via a dropping funnel. After the addition is completed, the temperature is kept at 5-10 °C for another 2 h. Subsequently, solvent is removed in vacuo and the reaction product is worked up by column chromatography (DCM). The title compound appears as a yellow solid (340 mg 47 %, HPLC purity 100.0 %, t_R = 15.0 min). 1H-NMR (400 MHz, CDCl_3) δ 9.09 (s, Ar-H, 1H), 8.73 (s, Ar-H, 1H), 5.48, (bs, CH-CH_3, 1H), 4.61 (bs, CH-CH_3, 1H), 1.95 (m, CH_2, 1H), 1.77 (bs, CH-CH_3, 4H), 1.66 (bs, CH-CH_3, 1H), 1.41 (bs, CH-CH_3, 6H) 13C NMR (101 MHz, CDCl_3) δ 166.2, 161.9, 144.0, 134.7, 133.2 (q, 3J_C,F = 3.5 Hz), 129.4 (q, 2J_C,F = 35.4 Hz), 126.9, 125.8 (q, 3J_C,F = 3.8 Hz), 122.5 (q, 1J_C,F = 273.6 Hz), 50.0, 49.0, 30.5, 29.76, 20.5, 19.9, 14.1. HRMS m/z calc for C_{16}H_{17}F_3N_3O_3S [M+H^+] 388.0938; found, 388.0933.

4-Hydroxypiperidin-1-thiourea (15a) Compound 15a is prepared by general procedure B, using 3-hydroxypiperidine (0.285 g, 2.82 mmol, 1 eq.) and thiocarbonyldiimidazole (0.580 g, 3.25 mmol, 1.15 eq.). The crude product is worked up column chromatographically (chloroform/MeOH 9:1) and the final compound is isolated as a white solid (206 mg 52 %). 1H-NMR (400 MHz, CDCl_3) δ 4.41 (m, cyclohexyl, 1H), 4.09 (m, cyclohexyl, 1H), 3.66 (m, cyclohexyl, 1H), 3.33 (m, cyclohexyl, 1H), 3.14 (m, cyclohexyl, 1H), 1.98 (m, cyclohexyl, 1H), 1.80 (m, cyclohexyl, 1H), 1.51 (m, cyclohexyl, 2H) MS m/z (ESI) 159.15 [M-H^+].

2-(4-Hydroxypiperidine-1-yl)-8-nitro-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (15b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (AR96) (0.168 g, 0.63 mmol, 1.0 eq.) by general procedure A. Compound 15b is prepared by general procedure C, using 4-hydroxypiperidine-1-thiourea
(15a) (0.100 g, 0.63 mmol, 1 eq.). The crude product is worked up by column chromatography (chloroform/MeOH 9:1) and recrystallised from EtAc/hexane (1:1). The title compound is isolated as a yellow solid (120 mg 51 %, HPLC purity 100.0 %, \( t_R = 12.5 \) min). \(^1\)H-NMR (400 MHz, CDCl\(_3\)) \( \delta 9.06 (d, \text{Ar-H}, J = 1.6 \text{ Hz}, 1\text{H}), 8.74 (s, \text{Ar-H}, 1\text{H}), 4.05 (m, \text{cyclohexyl, OH}, 5\text{H}), \)
\[ \text{2.66 (s, CH, 1H), 2.02 (m, cyclohexyl, 2H), 1.82 (m, cyclohexyl, 1H), 1.68 (m, cyclohexyl, 1H)} \]
\(^13\)C-NMR (100 MHz, CDCl\(_3\)) \( \delta 166.7, 162.9, 143.9, 134.3, 133.2, 129.6 (q, J_{C,F} = 35.9 \text{ Hz}), 126.5, 126.0, 122.3 (q, J_{C,F} = 273.1 \text{ Hz}), 65.9, 52.9, 47.5, 31.9, 22.0 \). HRMS \( m/z \) calc for C\(_{14}\)H\(_{13}\)F\(_3\)N\(_3\)O\(_4\)S [M+H\(^+\)], 376.0575; found, 376.0575.

**Thiomorpholin-4-thiourea (16a)** Dried NaSCN (389 mg, 4.80 mmol, 1.0 eq.) is suspended in acetone (10 ml), placed under argon and cooled to 5 °C. Benzoyl chloride (0.675 ml, 4.80 mmol, 1.0 eq.) is added slowly and the reaction mixture is stirred for 3 h at 5 °C. Thiomorpholine (500 mg, 4.80 mmol, 1.0 eq.) dissolved in acetone (5 ml) is then added at 12 °C, after which the reaction mixture is stirred at RT for 4 h. After the reaction, the solvent is removed in vacuo and the residue is dissolved in 36 % HCl (7.0 ml) and heated to 95 °C for 1.5 h. The aqueous solution is then mixed with acetone (7.0 ml) and solution is then neutralised with ammonia 36 % and extracted three times with DCM. The organic phases are collected, dried with MgSO\(_4\) and the evaporated on a rotary evaporator. The crude product is purified by column chromatography (EtAc/Heptane 1:1). The title compound is a white solid (270 mg 35 %). \(^1\)H-NMR (400 MHz, CDCl\(_3\)) \( \delta 6.48 (bs, \text{NH}_2, 2\text{H}), 4.11 (s, \text{CH}_2-N-\text{CH}_2, 4\text{H}), 2.74 (m, \text{CH}_2-S-\text{CH}_2, 4\text{H}) \) MS \( m/z \) (APCI) 163.0 [M+H\(^+\)].

**8-Nitro-2-(thiomorpholin-4-yl)-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (16b)** 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.332 g, 1.23 mmol, 1.0 eq.) by general procedure A. 16b is synthesised by general procedure C, using thiomorpholine-4-thiourea (16a) (0.200 g, 1.23
mmol, 1.0 eq.). The crude product is purified by recrystallisation from acetone (260 mg 56 %).

$^1$H-NMR (400 MHz, CDCl$_3$) $\delta$ 9.10 (d, Ar-H, $^4J = 1.6$ Hz, 1H), 8.77 (d, Ar-H, $^4J = 1.6$ Hz, 1H), 4.31 (bs, CH$_2$-N-CH$_2$, 4H), 2.81 (m, CH$_2$-S-CH$_2$, 4H) MS m/z (APCI) 378.1 [M+H$^+$].

8-Nitro-2-(1-oxo-1$\lambda^4$,4-thiomorpholin-4-yl)-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (16c) NaIO$_4$ (0.06 mg, 0.28 mmol, 1.05 eq.) is dissolved in 15 ml water and cooled to 0 °C with the aid of an ice bath. Now 8-nitro-2-(thiomorpholin-4-yl)-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (16b) dissolved in 5 ml MeOH is added, the reaction mixture is left at 0 °C for 4 h and then kept in the refrigerator for 65 h. Subsequently the aqueous phase is extracted three times with chloroform, the organic phases are collected, dried with MgSO$_4$ and the solvent is removed in vacuo. The crude product is purified by column chromatography (chloroform/MeOH 95:5). The title compound appears as a yellow solid (67 mg 64 %, HPLC purity 99.9 %, $t_R = 11.0$ min). $^1$H-NMR (400 MHz, CDCl$_3$) $\delta$ 9.11 (d, Ar-H, $^4J = 2.2$ Hz, 1H), 8.80 (d, Ar-H, $^4J = 2.2$ Hz, 1H), 4.59 (bs, CH$_2$-N-CH$_2$, 4H), 3.04 (m, CH$_2$-S-CH$_2$, 2H), 2.81 (m, CH$_2$-S-CH$_2$, 2H) $^{13}$C-NMR (100 MHz, CDCl$_3$): $\delta = 166.3$, 162.4, 144.0, 133.6 (q, $^2J_{CF} = 3.8$ Hz), 133.3, 130.3 (q, $^2J_{CF} = 35.3$ Hz), 126.8, 126.4 (q, $^3J_{CF} = 3.8$ Hz), 121.4 (q, $^1J_{CF} = 273.7$ Hz), 45.4, 37.4. HRMS m/z calc for C$_{13}$H$_{11}$F$_3$N$_3$O$_4$S$_2$ [M+H$^+$], 394.0139; found, 394.0143, m/z calc for C$_{13}$H$_{10}$F$_3$N$_3$O$_4$S$_2$Na [M+Na$^+$] 415.9958; found, 415.9958.

4-[8-Nitro-4-oxo-6-(trifluoromethyl)-4H-1,3-benzothiazin-2-yl]-1$\lambda^6$,4-thiomorpholin-1,1-dione (16d) 8-Nitro-2-(1-oxo-1$\lambda^4$,4-thiomorpholin-4-yl)-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (16b) (0.100 mg, 0.265 mmol, 1.0 eq.) is dissolved in 5 ml DCM and cooled to 0 °C. A solution of meta-chloroperbenzoic acid (m-CPBA) 65 % (0.165 mg, 0.624 mmol, 2.4 eq.) in 5 ml DCM is slowly added, the reaction mixture is slowly warmed to RT and left overnight at RT. Solvent is then removed under vacuum, the residue is dissolved in EtAc and washed twice with NaHCO$_3$ solution. The organic phase is dried over MgSO$_4$ and concentrated in vacuo.
Purification is carried out by recrystallisation from diisopropyl ether. The title compound appears as a white solid (55 mg 51%, HPLC purity 99.8%, t_R = 10.8 min). ^1H-NMR (400 MHz, CDCl_3) δ 9.12 (d, Ar-\textit{H}, J = 2.0 Hz, 1H), 8.83 (d, Ar-\textit{H}, J = 2.0 Hz, 1H), 4.52 (bs, CH_2-N-CH_2, 4H), 3.23 (m, CH_2-S-CH_2, 4H) ^13C-NMR (125 MHz, Aceton D_6) δ 165.4, 163.1, 144.7, 134.2, 131.9 (q, J_{CF} = 3.8 Hz), 128.7 (q, J_{CF} = 35.3 Hz), 126.7, 126.1 (q, J_{CF} = 3.8 Hz), 122.7 (q, J_{CF} = 271.8 Hz), 51.1, 44.6. HRMS m/z calc for C_{13}H_{10}F_{3}N_{3}O_{5}S_2 Na [M+Na]^+, 431.9908; found, 431.9906.

3-Hydroxyazetidine-1-thiourea (17a) Compound 17a is prepared according general procedure B, using 3-hydroxyazetidine\*HCl (0.100 g, 0.92 mmol, 1 eq.) and thiocarbonyldiimidazole (0.189 g, 1.06 mmol, 1.15 eq.). The crude product is purified by column chromatography (chloroform/MeOH 95:5). The title compound is a white solid (45 mg 37%) ^1H-NMR (500 MHz, CD_3OD) δ 4.51 (m, CH, 1H), 4.27 (bs, CH_2, 2H), 3.85 (bs, CH_2, 2H) MS m/z (APCI) 133.1 [M+H]^+.

2-(3-hydroxyazetidin-1-yl)-8-nitro-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (17b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.045 g, 0.16 mmol, 1.1 eq.) by general procedure A. 17b is prepared by general procedure C, using 3-hydroxyazetidine-1-thiourea (17a) (0.020 g, 0.15 mmol, 1 eq.). The crude product is purified by column chromatography (chloroform/MeOH 9:1). The title compound appears as a yellow solid (18 mg 35%, HPLC purity 99.2%, t_R = 14.9 min). ^1H-NMR (400 MHz, CD_3OD) δ 8.99 (s, Ar-\textit{H}, 1H), 8.87 (s, Ar-\textit{H}, 1H), 4.78 (bs, CH, 1H), 4.60 (bs, CH_2, 2H), 4.14 (m, CH_2, 2H) ^13C-NMR (100 MHz, CD_3OD) δ 166.7, 162.9, 144.0, 134.6, 132.1, 128.7 (q, J_{CF} = 35.3 Hz), 126.2, 125.8, 122.7 (q, J_{CF} = 271.8 Hz), 61.7, 61.5, 60.0 HRMS m/z calc for C_{12}H_{9}F_{3}N_{3}O_{4}S [M+H]^+, 348.0262; found, 348.0260, calc for C_{12}H_{10}F_{3}N_{3}O_{4}SNa [M+Na]^+, 370.0081; found, 370.0079.

Morpholine-4-thiourea (18a)
Synthesis by procedure B Compound 18a is prepared by general procedure B, using morpholine (0.450 g, 5.17 mmol, 1 eq.) and thiocarbonyldiimidazole (1.058 g, 5.94 mmol, 1.15 eq.). The crude product is purified by recrystallisation from isopropanol/diisopropyl ether (1:1). The title compound is isolated as a white solid (515 mg 68 %).

Synthesis by procedure E Synthesis according to general procedure E, starting from morpholine (4.35 ml, 50 mmol, 1 eq). Work-up after neutralization: the mixture was kept at 8 °C for 48 h, the precipitate filtered off, washed with a small amount of chloroform and dried. The title compound is isolated as a white solid (1.09 g, 15 %).

Synthesis by procedure F Synthesis according to general procedure F, starting from morpholine (166 mg, 1.91 mmol, 1 eq.). Compound 18a was isolated as a slightly brownish solid (115 mg, 41 %)

1H-NMR (400 MHz, CD3OD) δ 3.80 (m, CH2, 4H), 3.67 (m, CH2, 4H) MS m/z (ESI) 146.98 [M+H+].

2-(Morpholin-4-yl)-8-nitro-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (18b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.050 g, 0.18 mmol, 1.1 eq.) by general procedure A. Compound 18b is prepared by general procedure C using morpholine-4-thiourea (18a) (0.034 g, 0.23 mmol, 1 eq.). The crude product is purified by column chromatography (chloroform/MeOH 9:1). The title compound appears as a yellow solid (49 mg 75 %, HPLC purity 100.0 %, tR = 12.4 min). 1H-NMR (500 MHz, CDCl3) δ 9.08 (d, 1H, Ar-H, 4J = 1.5 Hz), 8.75 (d, 1H, Ar-H, 4J = 1.5 Hz), 4.01 (m, 4H, CH2-N-CH2), 3.82 (m, 4H, CH2-O-CH2) 13C-NMR (125 MHz, CDCl3) δ 166.3, 162.6, 143.9, 133.7, 133.5 (q, 3JC,F = 3.7 Hz), 129.9 (q, 2JC,F = 35.9 Hz), 126.8, 126.1 (q, 3JC,F = 3.7 Hz), 122.3 (q, 2JC,F = 272. 5 Hz), 66.3, 46.7 HRMS m/z calc for C13H10F3N3O4SNa [M+Na+] 384.0237; found, 384.0239.
4-[4-(Trifluoromethoxy)phenoxo]piperidine-1-thiourea (19a) Compound 19a is prepared by general procedure B, using 4-[4-(trifluoromethoxy)phenoxo]piperidine (0.100 g, 0.38 mmol, 1 eq.) and thiocarbonyldiimidazole (0.078 g, 0.44 mmol, 1.15 eq.). The crude product is purified by column chromatography (chloroform/MeOH 9:1). The title compound is a white solid (68 mg 56%). 1H-NMR (400 MHz, CD3OD) δ 7.18 (m, Ar-H, 2H), 7.03 (m, Ar-H, 2H), 4.65 (m, C-H, 1H), 4.05 (m, cyclohexyl, 2H), 3.82 (m, cyclohexyl, 2H), 2.01 (m, cyclohexyl, 2H), 1.78 (m, cyclohexyl, 2H) MS m/z (ESI) 319.15 [M-H]+.

8-Nitro-2-[4-[4-(trifluoromethoxy)phenoxo]piperidin-1-yl]-6-(trifluoromethyl)-4H-1,3-benzoiazin-4-one (19b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.049 g, 0.18 mmol, 1.1 eq.) by general procedure A. 19b is prepared by general procedure C, using 4-[4-(trifluoromethoxy)phenoxo]piperidine-1-thiourea (19a) (0.050 g, 0.16 mmol, 1 eq.). The crude product is purified by column chromatography (chloroform/MeOH 95:5). The title compound is a beige coloured solid (52 mg 60%, HPLC purity 100.0%, tR = 13.9 min). 1H-NMR (400 MHz, CDCl₃) δ 9.10 (d, Ar-H, 4J = 1.6 Hz, 1H), 8.75 (d, Ar-H, 4J = 1.6 Hz, 1H), 7.17 (m, Ar-H, 2H), 6.93 (m, Ar-H, 2H) 4.68 (m, C-H, 1H), 4.51 (bs, cyclohexyl, 1H), 3.98 (m, cyclohexyl, 3H), 2.06 (s, cyclohexyl, 4H) 13C-NMR (100 MHz, CDCl₃) δ 166.5, 161.9, 155.2, 143.9, 143.3, 134.0, 133.4, 129.7 (q, 2JCF = 36.4 Hz), 126.7, 126.0, 122.4 (q, 1JC,F = 272.8 Hz), 120.5 (q, 1JC,F = 255.6 Hz), 116.8, 70.8, 42.9, 30.3 HRMS m/z calc for C₂₁H₁₆F₆N₃O₅S [M+H]+, 536.0711; found, 536.0707, calc for C₂₁H₁₅F₆N₃O₅SNa [M+Na]+, 558.0527; found, 558.0534.

6-Methoxy-1,2,3,4-tetrahydroisoquinoline-2-thiourea (20a) Compound 20a is prepared by general procedure B, using 6-methoxy-1,2,3,4-tetrahydroisoquinoline (0.358 g, 2.20 mmol, 1 eq.) and thiocarbonyldiimidazole (0.450 g, 2.52 mmol, 1.15 eq.). The crude product is purified by column chromatography (LM chloroform/MeOH 95:5) and title compound appears as a
white solid (339 mg 62 %). $^1$H-NMR (400 MHz, CD$_3$OD) $\delta$ 7.08 (m, Ar-H, 1H), 6.79 (m, Ar-H, 2H), 4.83 (m, CH$_2$, 2H, close to solvent signal), 3.95 (m, CH$_2$, 2H), 3.79 (s, CH$_3$, 3H), 2.91 (t, CH$_2$, $^2$J = 5.9 Hz, 2H) MS m/z (ESI) 223.06 [M+H$^+$].

2-(6-Methoxy-1,2,3,4-tetrahydroisoquinolin-2-yl)-8-nitro-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (20b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.399 g, 1.49 mmol, 1.1 eq.) by general procedure A. Compound 20b is prepared by general method 3, using 6-methoxy-1,2,3,4-tetrahydroisoquinoline-2-thiourea (20a) (0.300 g, 1.35 mmol, 1 eq.). The crude product is purified by column chromatography (chloroform/MeOH 99:1). The title compound is a yellow solid (545 mg 92 %, HPLC purity 95.2 %, $t_R = 15.0$ min). $^1$H-NMR (400 MHz, CDCl$_3$) $\delta$ 9.13 (d, Ar-H, $^4$J = 1.6 Hz, 1H), 8.76 (d, Ar-H, $^4$J = 1.6 Hz, 1H), 7.14 (d, Ar-H, $^3$J = 8.4 Hz, 1H), 6.81 (m, Ar-H, 1H), 6.75 (s, Ar-H, 1H), 5.01 (m, CH$_2$, 2H), 4.15 (m, CH$_2$, 2H), 3.80 (s, CH$_3$, 3H), 3.02 (m, CH$_2$, 2H) $^{13}$C-NMR (100 MHz, CDCl$_3$) $\delta$ 166.1, 162.3, 159.0, 143.9, 134.2, 133.5, 129.7 (q, $^2$J$_{CF}$ = 36.2 Hz), 127.7, 126.9, 126.0, 122.4 (q, $^1$J$_{CF}$ = 272.8 Hz), 113.0, 55.4, 47.8, 44.5, 29.0 HRMS m/z calc for C$_{19}$H$_{15}$F$_3$N$_3$O$_4$S [M+H$^+$], 438.0731; found, 438.0735, calc for C$_{19}$H$_{14}$F$_3$N$_3$O$_4$SNa [M+Na$^+$], 460.0551; found, 460.0553.

7-Methoxy-1,2,3,4-tetrahydroisoquinoline-2-thiourea (21a) Compound 21a is prepared by general procedure B, using 7-methoxy-1,2,3,4-tetrahydroisoquinoline (0.073 g, 0.45 mmol, 1 eq.) and thiocarbonyldiimidazole (0.092 g, 0.51 mmol, 1.15 eq.). The crude product is purified by column chromatography (LM chloroform/MeOH/ammonia 36 % 98:2:1). The title compound appears as a white solid (76 mg 76 %). $^1$H-NMR (400 MHz, CDCl$_3$) $\delta$ 7.08 (d, Ar-H, $^3$J = 8.4 Hz, 1H), 6.77 (dd, Ar-H, $^3$J = 8.2 Hz, $^4$J = 2.5 Hz, 1H), 6.71 (d, Ar-H, $^4$J = 2.5 Hz, 1H), 5.95 (bs, NH$_2$, 2H), 4.89 (bs, CH$_2$, 2H), 3.90 (bs, CH$_2$, 2H), 3.77 (s, CH$_3$, 3H), 2.88 (t, CH$_2$, $^3$J = 5.9 Hz, 2H) MS m/z (ESI) 221.25 [M-H$^+$].
2-(7-Methoxy-1,2,3,4-tetrahydroisoquinolin-2-yl)-8-nitro-6-(trifluoromethyl)-4H-1,3-
benzothiazin-4-one (21b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is
prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.026 g, 0.10 mmol, 1.1 eq.)
by general procedure A. Compound 21b is prepared by general procedure C, using 7-methoxy-
1,2,3,4-tetrahydroisoquinoline-2-thiourea (21a) (0.020 g, 0.09 mmol, 1 eq.). The crude
product is worked up by column chromatography (chloroform/MeOH 98:2) and recrystallised
from EtAc/hexane (1:1). The title compound appears as a yellow solid (14 mg 36 %, HPLC
purity 100.0 %, tR = 15.1 min). 1H-NMR (400 MHz, CDCl3) δ 9.14 (s, Ar-H, 1H), 8.77 (s, Ar-H, 1H),
7.13 (d, Ar-H, 3J = 8.4 Hz, 1H), 6.82 (d, Ar-H, 3J = 8.4 Hz, 1H), 6.77 (s, Ar-H, 1H), 5.05 (m, CH2,
2H), 4.16 (m, CH3, 2H), 3.81 (s, CH3, 3H), 3.00 (m, CH2, 2H) 13C-NMR (100 MHz, CDCl3) δ 166.1,
162.4, 158.7, 143.9, 134.1, 133.6 (q, 3JCF = 3.8 Hz), 129.8 (q, 2JCF = 35.1 Hz), 129.0, 126.9, 126.3,
126.0 (q, 3JCF = 3.8 Hz), 121.4 (q, 1JCF = 273.9 Hz), 113.8, 111.3, 55.4, 48.4, 45.0, 28.0 0 HRMS
m/z calc for C19H15F3N3O4S [M+H+], 438.0731; found, 438.0731.

6,7-Dimethoxy-1,2,3,4-tetrahydroisoquinoline-2-thiourea (22a) Compound 22a is prepared by
general procedure B, using 6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline (0.721 g, 3.73 mmol,
1.0 eq.) and thiocarboxyldiimidazole (0.765 g, 4.29 mmol, 1.15 eq.). The crude product
is purified by column chromatography (chloroform/MeOH/ammonia 36 % 98:2:1). The title compound appears as a white solid (218 mg 23 %) 1H-NMR (400 MHz, CD3OD) δ 6.78 (s, Ar-H,
1H), 6.74 (s, Ar-H, 1H), 4.84 (bs, N-CH2, 2H, Wasser Signal), 3.95 (m, N-CH2, 2H), 3.80 (s, O-CH3,
3H), 3.79 (s, O-CH3, 3H), 2.84 (t, CH2, 3J = 5.9 Hz, 2H) MS m/z (APCI) 253.1 [M+H+].

2-(6,7-Dimethoxy-1,2,3,4-tetrahydroisoquinolin-2-yl)-8-nitro-6-(trifluoromethyl)-4H-1,3-
benzothiazin-4-one (22b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is
prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.229 g, 0.85 mmol, 1.1 eq.)
by general procedure A. Compound 22b is synthesised by general procedure C, using 6,7-
dimethoxy-1,2,3,4-tetrahydroisoquinoline-2-thiourea (22a) (0.194 g, 0.77 mmol, 1.0 eq.). The crude product is purified by column chromatographically (chloroform) and re-crystallised from EtOH. The title compound appears as a yellow solid (137 mg 38 %, HPLC purity 97.7 %, \( t_R = 14.5 \) min). \(^1\)H-NMR (400 MHz, CDCl\(_3\)) \( \delta \) 9.13 (d, Ar-H, \( ^4J = 1.6 \) Hz, 1H), 8.77 (d, Ar-H, \( ^4J = 1.6 \) Hz, 1H), 6.71 (s, Ar-H, 1H), 6.68 (s, Ar-H, 1H), 5.02 (m, N-CH\(_2\), 2H), 4.19 (m, N-CH\(_2\), 2H), 3.87 (s, O-C\(_3\)H\(_3\), 6H), 2.98 (m, CH\(_2\), 2H) \(^{13}\)C-NMR (125 MHz, CDCl\(_3\)) \( \delta \) 166.2, 162.2, 148.3, 143.9, 134.2, 133.5 (q, \( ^3J_{CF} = 3.8 \) Hz), 129.8 (q, \( ^2J_{CF} = 35.8 \) Hz), 126.9, 126.0 (q, \( ^3J_{CF} = 3.8 \) Hz), 125.3, 123.8, 122.4 (q, \( ^1J_{CF} = 273.1 \) Hz), 111.2, 109.4, 56.2, 56.0, 48.0, 44.7, 28.1 HRMS m/z calc for C\(_{20}\)H\(_{17}\)F\(_3\)N\(_3\)O\(_5\)S [M+H\(^+\)], 468.0837; found, 468.0841, calc for C\(_{20}\)H\(_{16}\)F\(_3\)N\(_3\)O\(_5\)SNa [M+Na\(^+\)], 490.0657; found, 490.0661.

1,2,3,4-Tetrahydroisoquinoline-2-thiourea (23a) Compound 23a is prepared by general procedure B, using 1,2,3,4-tetrahydroisoquinoline (0.750 g, 5.63 mmol, 1.0 eq.) and thiocarbonyldiimidazole (1.154 g, 6.48 mmol, 1.15 eq.). The crude product is recrystallised from diisopropyl ether/isopropanol 1:1. The title compound appears as a white solid (610 mg 56 %) \(^1\)H-NMR (400 MHz, CD\(_3\)OD) \( \delta \) 7.18 (m, Ar-H, 4H), 4.89 (bs, N-CH\(_2\), 2H), 3.95 (bs, N-CH\(_2\), 2H), 2.91 (t, CH\(_2\), \( ^3J = 5.9 \) Hz, 2H) MS m/z (ESI) 191.21 [M-H\(^-\)].

8-Nitro-2-(1,2,3,4-tetrahydroisoquinolin-2-yl)-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (23b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.463 g, 1.72 mmol, 1.1 eq.) by general procedure a. 23b is synthesised by general procedure C, using 1,2,3,4-tetrahydroisoquinoline-2-thiourea (23a) (0.300 g, 1.56 mmol, 1.0 eq.). The crude product is purified by column chromatography (chloroform). The final product appears as a yellow solid (438 mg 69 %, HPLC purity 99.1 %, \( t_R = 15.0 \) min). \(^1\)H-NMR (400 MHz, CDCl\(_3\)) \( \delta \) 9.13 (d, Ar-H, \( ^4J = 2.0 \) Hz, 1H), 8.77 (d, Ar-H, \( ^4J = 2.0 \) Hz, 1H), 7.25 (m, Ar-H, 4H), 5.08 (m, N-CH\(_2\), 2H), 4.17 (m, N-CH\(_2\), 2H), 3.07 (m, CH\(_2\), 2H) \(^{13}\)C-
NMR (125 MHz, CDCl₃) δ 166.1, 162.3, 143.9, 134.6, 134.1, 133.7, 133.5 (q, ²J_C,F = 3.8 Hz), 131.8, 129.7 (q, ²J_C,F = 35.8 Hz), 128.7, 128.0, 127.7, 127.2, 126.6, 126.8, 126.0 (q, ²J_C,F = 3.8 Hz), 122.4 (q, ²J_C,F = 273.1 Hz), 48.2, 44.6, 28.7

HRMS m/z calc for C₁₈H₁₂F₃N₃O₃SNa [M+Na⁺], 430.0445; found, 430.0438.

4-(Piperidin-1-yl)piperidine-1-thiourea (24a) Compound 24a is prepared by general procedure B, using 4-(piperidin-1-yl)piperidine (0.100 g, 0.59 mmol, 1 eq.) and thiocarbonyldiimidazole (0.121 g, 0.68 mmol, 1.15 eq.). The crude product is purified by column chromatography (chloroform/MeOH/ammonia 36 % 90:10:1). The title compound appears as a white solid (24 mg 18 %) ¹H-NMR (400 MHz, CDCl₃) δ 5.76 (s, NH₂, 2H), 4.53 (s, CH, 1H), 3.06 (m, cyclohexyl, 2H), 2.51 (m, cyclohexyl, 5H), 1.88 (m, cyclohexyl, 2H), 1.60 (m, cyclohexyl, 7H), 1.43 (m, cyclohexyl, 2H) MS m/z (ESI) 228.13 [M+H⁺].

8-Nitro-2-[4-(piperidin-1-yl)piperidin-1-yl]-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (24b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.024 g, 0.09 mmol, 1.1 eq.) by general procedure A. 24b is prepared by general procedure C, using 4-(piperidin-1-yl)piperidin-1-thiourea (24a) (0.018 g, 0.08 mmol, 1 eq.). The crude product is worked up by column chromatography (chloroform/MeOH/ammonia 36 % 98:2:1). The title compound appears as a yellow solid (20 mg 57 %, HPLC purity 99.4 %, tᵦ = 12.4 min) ¹H-NMR (400 MHz, CDCl₃) δ 9.10 (d, Ar-H, ²J = 1.6 Hz, 1H), 8.74 (d, Ar-H, ²J = 1.6 Hz, 1H), 5.22 (bs, cyclohexyl, 1H), 4.42 (bs, cyclohexyl, 1H), 3.18 (m, cyclohexyl, 2H), 2.65 (m, CH, 1H), 2.52 (m, cyclohexyl, 4H), 2.03 (m, cyclohexyl, 2H), 1.63 (m, cyclohexyl, 6H), 1.45 (m, cyclohexyl, 2H) ¹³C-NMR (100 MHz, CDCl₃) δ 166.5, 161.6, 143.9, 134.2, 133.3, 129.6 (q, ²J_C,F = 35.1 Hz), 126.7, 125.9, 126.0, 122.4 (q, ²J_C,F = 273.1 Hz), 61.8, 50.3, 46.1 28.1 26.2, 24.5 HRMS m/z calc for C₁₉H₂₁F₃N₄O₃SNa [M+Na⁺], 443.1361; found, 443.1357, calc for C₁₉H₂₁F₃N₄O₃SNa [M+Na⁺], 465.1180; found, 465.1176.
5-Benzyl-octahydropyrrolo[3,4-c]pyrrole-2-thiourea (25a) Compound 25a is prepared by general procedure B, using 2-benzyl-octahydropyrrolo[3,4-c]pyrrole (0.100 g, 0.49 mmol, 1 eq.) and thiocarbonyldiimidazole (0.101 g, 0.57 mmol, 1.15 eq.). The crude product is purified by column chromatography (chloroform/MeOH 98:2) and the title compound is a white solid (58 mg 45%). ¹H-NMR (400 MHz, CDCl₃) δ 7.28 (m, Ar-H, 5H), 5.67 (bs, NH₂, 2H), 3.87 (bs, Octahydropyrrolo[3,4-c]pyrrole, 2H), 3.59 (s, CH₂, 2H), 3.42 (bs, octahydropyrrolo[3,4-c]pyrrole, 2H), 2.94 (bs, octahydropyrrolo[3,4-c]pyrrole, 2H), 2.60 (m, octahydropyrrolo[3,4-c]pyrrole, 2H), 2.53 (m, octahydropyrrolo[3,4-c]pyrrole, 2H) MS m/z (ESI) 262.12 [M+H⁺].

2-(5-Benzyl-octahydropyrrolo[3,4-c]pyrrol-2-yl)-8-nitro-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (25b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.045 g, 0.17 mmol, 1.1 eq.) by general procedure A. 25b is prepared by general procedure C, using 5-benzyl-octahydropyrrolo[3,4-c]pyrrole-2-thiourea (25a) (0.040 g, 0.15 mmol, 1 eq.). The crude product is purified by column chromatography (chloroform/MeOH 98:2). The title compound appears as a light yellow solid (50 mg 69%, HPLC purity 94.9%, tᵣ = 12.9 min) ¹H-NMR (400 MHz, CDCl₃) δ 9.15 (s, Ar-H, 1H), 8.76 (s, Ar-H, 1H), 7.28 (m, Ar-H, 5H), 4.18 (m, Octahydropyrrolo[3,4-c]pyrrole, 1H), 3.94 (m, Octahydropyrrolo[3,4-c]pyrrole, 2H), 3.62 (m, Octahydropyrrolo[3,4-c]pyrrole, 1H), 3.11 (bs, Octahydropyrrolo[3,4-c]pyrrole, 1H), 2.99 (bs, Octahydropyrrolo[3,4-c]pyrrole, 1H), 2.70 (m, Octahydropyrrolo[3,4-c]pyrrole, 2H), 2.58 (m, Octahydropyrrolo[3,4-c]pyrrole, 2H) ¹⁳C-NMR (100 MHz, CDCl₃) δ 165.9, 159.8, 143.6, 138.4, 134.6, 133.7 (q, ³J₇C,F = 3.8 Hz), 129.6 (q, ³J₇C,F = 35.8 Hz), 128.5, 128.4, 127.2, 126.8, 125.8 (q, ³J₇C,F = 3.8 Hz), 122.4 (q, ³J₇C,F = 273.1 Hz), 59.7, 59.6, 59.1, 56.3, 53.3, 41.8, 40.0 HRMS m/z calc for C₂₂H₂₀F₃N₄O₃S [M+H⁺], 477.1204; found, 477.1206.
tert-Butyl 6-thiourea-octahydro-1H-pyrrolo[3,4-b]pyridine-1-carboxylate (26a) Compound 26a is prepared by general procedure B, using tert-butyloctahydro-1H-pyrrolo[3,4-b]pyridine-1-carboxylate (0.500 g, 2.21 mmol, 1.0 eq.) and thiocarbonyldiimidazole (0.453 g, 2.54 mmol, 1.15 eq.). For better solubility of tert-butyloctahydro-1H-pyrrolo[3,4-b]pyridine-1-carboxylate 3 ml MeOH are added to the reaction mixture. The crude product is purified by column chromatography (EtAc). The title compound appears as a white solid (397 mg 63 %). 1H-NMR (400 MHz, CD3OD) δ 4.69 (bs, 2,8-diazabicyclo[4.3.0]nonane, 1H), 3.90 (m, 2,8-diazabicyclo[4.3.0]nonane, 2H), 3.56 (m, 2,8-diazabicyclo[4.3.0]nonane, 2H), 3.35 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 2.81 (bs, 2,8-diazabicyclo[4.3.0]nonane, 1H), 2.26 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 1.36 (m, 2,8-diazabicyclo[4.3.0]nonane, 2H) MS m/z (ESI) 285.99 [M+H]+, 303.03 [M+Na]+.

tert-Butyl 6-[8-nitro-4-oxo-6-(trifluoromethyl)-4H-1,3-benzothiazin-2-yl]-octahydro-1H-pyrrolo[3,4-b]pyridine-1-carboxylate (26b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.215 g, 0.80 mmol, 1.1 eq.) by general procedure A. 26b is synthesised by general procedure C, using tert-butyl-6-thiourea octahydro-1H-pyrrolo[3,4-b]pyridine-1-carboxylate (26a) (0.205 g, 0.72 mmol, 1.0 eq.). The crude product is purified by column chromatography (EtAc/heptane 1:1). The title compound appears as a yellow solid (180 mg 50 %, HPLC purity 98.6 %, tR = 15.0 min). 1H-NMR (400 MHz, CDCl3) δ 9.06 (m, Ar-H, 1H), 8.69 (d, Ar-H, J = 1.7 Hz, 1H), 4.83 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 4.02 (m, 2,8-diazabicyclo[4.3.0]nonane, 2H), 3.77 (m, 2,8-diazabicyclo[4.3.0]nonane, 0.5H), 3.72 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 3.58 (m, 2,8-diazabicyclo[4.3.0]nonane, 0.5H), 3.48 (m, 2,8-diazabicyclo[4.3.0]nonane, 0.5H), 2.71 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 2.34 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 1.70 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 1.44 (s, t-Boc,
(26c) tert-butyl 6-[8-nitro-4-oxo-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one] octahydro-1H-pyrrolo[3,4-b]pyridine-1-carboxylate (26b) (0.273 g, 0.55 mmol, 1.0 eq.) is dissolved in a mixture of 2 ml DCM and 2 ml TFA and stirred at room temperature for 1.5 h. The solvent is removed under vacuum and the reaction mixture is co-evaporated three times each with toluene and chloroform. The crude product purified by column chromatography (chloroform/MeOH/ammonia 36 % 95:5:1). The title compound appears as a yellow amorphous substance (164 mg 75 %, HPLC purity 96.0 %, \(t_R = 13.5\) min). \(^1\)H-NMR (400 MHz, CDCl\(_3\)) 8 9.05 (m, Ar-H, 1H), 8.66 (m, Ar-H, 1H), 3.71 (m, 2,8-diazabicyclo[4.3.0]nonane, 4H), 3.40 (m, NH, 1H), 2.94 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 2.62 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 2.52 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 2.39 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 1.68 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H) \(^1\)C-NMR (100 MHz, CDCl\(_3\)) 8 165.7, 161.0, 143.4, 134.1, 133.5, 129.4, 126.8, 125.7, 122.4 (q, \(^1\)J\(_{CF}\) = 273.1 Hz), 57.9, 56.1, 54.9, 54.6, 51.4, 48.1, 45.2, 44.9, 37.3, 35.5, 22.8, 22.7, 21.1, 20.8 HRMS m/z calc for C\(_{21}\)H\(_{24}\)F\(_3\)N\(_4\)O\(_5\)S [M+H\(^+\)], 501.1415; found, 501.1411.

8-Nitro-2-{octahydro-1H-pyrrolo[3,4-b]pyridin-6-yl]-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (26d) 8-Nitro-2-{octahydro-1H-pyrrolo[3,4-b]pyridin-6-yl]-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (26c) (0.020 g, 0.05 mmol, 1.0 eq.) is dissolved in a mixture of equal parts DCM and saturated aqueous Na\(_2\)CO\(_3\) solution. To this solution is added benzyl bromide (0.011 g, 0.06 mmol, 1.24 eq.) and the reaction mixture is left at room
temperature for 12 h. The aqueous phase is then extracted three times with DCM. The organic phases are collected, dried with MgSO₄ and the solvent is removed in vacuo. Further purification is carried out by column chromatography (chloroform/ammonia 36 % 99:1). The title compound as a yellow solid (18 mg 72 %, HPLC purity 99.5%, tᵣ = 12.4 min). ¹H-NMR (400 MHz, CDCl₃) δ 9.17 (m, Ar-H, 1H), 8.76 (m, Ar-H, 1H), 7.25 (m, Ar-H, 5H), 3.82 (m, 2,8-diazabicyclo[4.3.0]nonane, N-CH₂-Ar, 4H), 3.51 (m, N-CH₂-Ar, 1H), 3.34 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 2.72 (bs, 2,8-diazabicyclo[4.3.0]nonane, 1H), 2.31 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 1.66 (m, 2,8-diazabicyclo[4.3.0]nonane, 5H) ¹³C-APT-NMR (100 MHz, CDCl₃) δ 165.9, 161.0, 143.6, 138.6, 134.5, 133.7, 129.5 (q, ²JCF = 35.9 Hz), 128.8, 128.4, 128.3, 128.2, 127.1, 126.9, 125.8, 122.4 (q, ²JCF = 273.1 Hz), 61.9, 60.2, 59.6, 59.5, 52.6, 51.6, 50.4, 50.0, 49.7, 48.4, 37.7, 36.1, 23.6, 22.1 HRMS m/z calc for C₂₃H₂₂F₃N₄O₃S [M+H⁺], 491.1361; found, 491.1346.

6-Benzyl-octahydro-1H-pyrrolo[3,4-b]pyridine-1-thiourea (27a) Compound 27a is prepared by general procedure B, using 6-benzyl-octahydro-1H-pyrrolo[3,4-b]pyridine (0.140 g, 0.64 mmol, 1.0 eq.) and thiocarbonyldiimidazole (0.131 g, 0.73 mmol, 1.15 eq.). The crude product is processed by column chromatography (chloroform/MeOH/ammonia 36 % 90:10:1). The title compound appears as a white solid (95 mg 54 %). ¹H-NMR (400 MHz, CDCl₃) δ 7.26 (m, Ar-H, 5H), 5.97 (bs, NH₂, 2H), 5.15 (bs, 2,8-diazabicyclo[4.3.0]nonane, 1H), 4.19 (bs, 2,8-diazabicyclo[4.3.0]nonane, 1H), 3.68 (d, N-CH₂-Ar, ²J = 13.1 Hz, 1H), 3.60 (d, N-CH₂-Ar, ²J = 13.1 Hz, 1H), 3.12 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 2.98 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 2.78 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 2.58 (t, 2,8-diazabicyclo[4.3.0]nonane, ³J = 6.8 Hz, 1H), 2.52 (dd, 2,8-diazabicyclo[4.3.0]nonane, ³J = 9.2 Hz, ⁴J = 2.0 Hz, 1H), 2.36 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 1.86 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 1.70 (m, 2,8-
diazabicyclo[4.3.0]nonane, 1H), 1.57 (m, 2,8-diazabicyclo[4.3.0]nonane, 2H) MS m/z (ESI) 276.10 [M+H⁺].

2-[(6-Benzyl-octahydro-1H-pyrrolo[3,4-b]pyridin-1-yl)-8-nitro-6-(trifluoromethyl)-4H-1,3-benzothiazin-4-one (27b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.102 g, 0.38 mmol, 1.1 eq.) by general method 1. 27b is synthesised by general procedure C, using tert-6-benzyl-octahydro-1H-pyrrolo[3,4-b]pyridine-1-thiourea (27a) (0.095 g, 0.34 mmol, 1.0 eq.). The crude product is purified by column chromatography (chloroform/ammonia 36 % 99:1). The title compound is a yellow solid (54 mg 32 %, HPLC purity 99.0 %, tᵣ = 14.1 min). ¹H-NMR (400 MHz, CDCl₃) δ 9.07 (d, Ar-H, 4J = 1.6 Hz, 1H), 8.72 (m, Ar-H, 4J = 1.6 Hz, 1H), 7.28 (m, Ar-H, 5H), 5.66 (bs, 2,8-diazabicyclo[4.3.0]nonane, 0.5H), 5.02 (bs, 2,8-diazabicyclo[4.3.0]nonane, 0.5H), 4.80 (bs, 2,8-diazabicyclo[4.3.0]nonane, 0.5H), 4.12 (bs, 2,8-diazabicyclo[4.3.0]nonane, 0.5H), 3.74 (bs, CH₂-Ar, 1H) 3.65 (m, CH₂-Ar, 1H), 3.41 (bs, 2,8-diazabicyclo[4.3.0]nonane, 0.5H), 3.09 (bs, 2,8-diazabicyclo[4.3.0]nonane, 0.5H), 3.02 (t, 2,8-diazabicyclo[4.3.0]nonane, 4J = 9.5 Hz, 1H), 2.69 (m, 2,8-diazabicyclo[4.3.0]nonane, 3H) 2.42 (m, 2,8-diazabicyclo[4.3.0]nonane, 1H), 1.94 (m, 2,8-diazabicyclo[4.3.0]nonane, 4H) ¹³C-NMR (125 MHz, CDCl₃) δ 166.2, 162.9, 143.9, 138.6, 134.3, 133.3 (q, 3J_C,F = 3.8 Hz), 129.6 (q, 3J_C,F = 35.3 Hz),128.5, 128.3, 127.1, 126.8, 125.9 (q, 3J_C,F = 3.8 Hz), 122.4 (q, 3J_C,F = 272.8 Hz), 60.0, 58.7, 56.1, 55.0, 54.4, 43.4, 36.4, 35.9, 26.4, 25.7, 22.9, 22.1 HRMS m/z calc for C₂₃H₂₂F₃N₄O₃S [M+H⁺], 491.1360; found, 491.1354.

1-(Cyclohexylmethyl)piperazine (28a) N-formylpiperazine (1.32 ml, 12.8 mmol, 1.0 eq.), cyclohexylmethyl bromide (1.97 ml, 14.1 mmol, 1.1 eq.), KI (0.026 g, 0.18 mmol, 0.014 eq.) and K₂CO₃ (2.13 g, 15.4 mmol, 1.2 eq.) are dissolved in a round bottom flask in 10 ml acetonitrile. After 23 h heating under reflux the reaction mixture is filtered and the filtrate is concentrated on the rotary evaporator. After dissolving the residue in 5 N NaOH solution (5.0
ml) and EtOH (9.3 ml), it is heated for 14 h under reflux. After the reaction is complete, the EtOH is removed under vacuum, distilled water is added to the residue and extracted three times with DCM. The organic layers are collected, dried over MgSO$_4$ and the solvent removed under vacuum. The crude product is purified by column chromatography chloroform/MeOH/ammonia 36 %. (9:1:0.1). The title compound is a colourless oil (1.83 g 79 %). $^1$H-NMR (400 MHz, CDCl$_3$) $\delta$ 3.76 (bs, N-H, 1H), 2.95 (t, CH$_2$-N-CH$_2$, $^3$J=4.9 Hz, 4H), 2.43 (bs, CH$_2$-N-CH$_2$, 4H), 2.11 (d, N-CH$_2$-CH, $^3$J=7.2 Hz, 2H), 1.70 (m, Cyclohexyl, 5H), 1.46 (m, CH$_2$-CH-(CH$_2$)$_2$, 1H), 1.18 (m, Cyclohexyl, 3H), 0.86 (m, Cyclohexyl, 2H) MS m/z (ESI) 183.30 [M+H$^+$].

4-(Cyclohexylmethyl)piperazin-1-thiourea (28b) Compound 28b is prepared by general procedure B, using 3-cyclohexylmethylpiperazine (28a) (0.190 g, 1.04 mmol, 1 eq.) and thiocarbonyldiimidazole (0.214 g, 1.20 mmol, 1.15 eq.). The crude product is purified by column chromatography (LM chloroform/MeOH 98:2). The title compound appears as a brownish solid (118 mg 52 %). $^1$H-NMR (400 MHz, CDCl$_3$) $\delta$ 5.85 (m, NH$_2$, 2H), 3.83 (m, CH$_2$-N-CH$_2$, 4H), 2.47 (m, CH$_2$-N-CH$_2$, 4H), 2.17 (d, N-CH$_2$-CH, $^3$J = 7.2 Hz, 2H), 1.71 (m, cyclohexyl, 5H), 1.47 (m, N-CH$_2$-CH, 1H), 1.19 (m, cyclohexyl, 3H), 0.88 (m, cyclohexyl, 2H) MS m/z (ESI): 242.12 [M+H$^+$].

Macozinone 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.282 g, 1.04 mmol, 1.0 eq.) according to general procedure A. The synthesis of the benzothiazinone is carried out according to general procedure C. The precipitated product is isolated by filtration and recrystallised from acetone. The title compound is light yellow solid (293 mg 68 %, HPLC purity 100.0 %, $t_R = 12.5$ min). $^1$H-NMR (400 MHz, CDCl$_3$) $\delta$ 9.09 (d, Ar-H, $^4$J = 2.0, 1H), 8.75 (d, Ar-H, $^4$J = 2.0, 1H), 3.98 (m, CH$_2$-N-CH$_2$, 4H), 2.56 (m, CH$_2$-N-CH$_2$, 4H), 2.20 (m, N-CH$_2$-CH, 2H), 1.74 (m, cyclohexyl, 5H), 1.51 (m, N-CH$_2$-CH, 1H), 1.22 (m, cyclohexyl, 3H), 0.89 (m, cyclohexyl, 2H) $^{13}$C-NMR (100 MHz,
CDCl$_3$ $\delta$ 166.4, 162.0, 143.9, 133.4 ($q$, $^3J_{CF}$ = 3.4 Hz), 129.7 ($q$, $^2J_{CF}$ = 35.5 Hz), 126.8, 126.0 ($q$, $^3J_{CF}$ = 3.8 Hz), 122.4 ($q$, $^1J_{CF}$ = 273.1 Hz), 65.1, 53.1, 46.6, 35.0, 31.7, 26.7, 26.0 HRMS m/z calc for C$_{23}$H$_{22}$F$_3$N$_4$O$_3$ [M+H$^+$], 457.15217; found, 457.1512.

(2S)-2-Methyl-1,4-dioxo-8-azaspiro[4.5]decane (29a) 4-Piperidone monohydrate (995 mg, 6.57 mmol, 1 eq.) and (2S)-propane-1,2-diol (0.500 mg, 6.57 mmol, 1 eq.) are dissolved in anisole (50 ml). After addition of p-toluenesulfonic acid monohydrate (0.125 mg, 0.66 mmol, 0.1 eq.) the reaction mixture is boiled for 4 h on the water separator. Then the anisole is removed on the rotary evaporator and the crude product obtained is suspended in brine. After addition of NaOH solution (2 N, pH 12) the product is extracted from the aqueous phase with diethyl ether (six times). The ether phases are combined, dried over MgSO$_4$ and the solvent removed in vacuo. The product appears as a colourless oil (186 mg 15 %). $^1$H-NMR (400 MHz, CDCl$_3$) $\delta$ 4.22 (m, CH$_2$-CH$_3$, 1H), 4.05 (dd, CH$_2$-CH$_3$, $^2J$ = 7.8 Hz, 1H), 3.44 (t, CH$_2$-CH$_3$, $^2J$ = 7.8 Hz, 1H), 2.92 (m, CH$_2$-N-CH$_2$, 4H), 1.87 (bs, N-H, 1H), 1.67 (m, CH$_2$-C-CH$_2$, 4H), 1.27 (d, CH$_3$, $^3J$ = 6.0 Hz, 3H) MS m/z (APCI) 158.2 [M+H$^+$].

(2S)-2-Methyl-1,4-dioxo-8-azaspiro[4.5]decane-thiourea (29b) Dried NaSCN (338 mg, 4.16 mmol, 1.0 eq.) is suspended in acetone (10 ml), placed under inert gas and cooled to 5 °C. Benzoyl chloride (0.480 ml, 4.16 mmol, 1.0 eq.) is added slowly and the reaction mixture is stirred for 3.5 h at 5 °C. This is followed by the addition of (2S)-2-methyl-1,4-dioxo-8-azaspiro[4.5]decane (29a) (186 mg, 4.16 mmol, 1.0 eq.) dissolved in acetone (5 ml) at 12 °C, then the reaction mixture is stirred at room temperature overnight. After the reaction, the solvent is removed under vacuum and the residue is dissolved in 19 ml MeOH/water mixture (3:1) and K$_2$CO$_3$ (1152 mg, 8.34 mmol, 1.0 eq.) is added. The reaction mixture is heated under reflux for 24 h. After the reaction is complete, the solid is removed by filtration and the solvent is concentrated in vacuo. The crude product is suspended in brine and the aqueous phase is
extracted with EtAc (three times). The organic layers are collected, dried (MgSO₄) and concentrated. The crude product is purified by column chromatography (TBME). The product is a white solid (111 mg 43 %). $^{1}$H-NMR (400 MHz, CDCl₃) δ 6.07 (bs, N-H, 2H), 4.23 (m, CH₂-CH-CH₃, 1H), 4.06 (dd, CH₂-CH-CH₃, $^2$J = 8.0 Hz, 1H), 3.88 (m, CH₂-N-CH₂, 4H), 3.44 (t, CH₂-CH-CH₃, $^2$J = 7.8 Hz, 1H), 1.76 (m, CH₂-C-CH₂, 4H), 1.26 (d, CH₃, $^3$J = 6.1 Hz, 3H) MS m/z (APCI) 217.1 [M+H⁺].

BTZ043 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.57 g, 0.21 mmol, 1.0 eq.) by general procedure A, replacing toluene with anisole. BTZ043 is synthesised by general procedure C in anisole in place of toluene, using (2S)-2-methyl-1,4-dioxa-8-azaspiro[4.5]decan-8-thiourea (29b) (0.47 g, 0.21 mmol, 1.0 eq.). The crude product is purified chromatographically (LM TBME). The title compound appears as a yellow solid (66 mg 73 %, HPLC purity 100.0 %, $t_R$ = 14.2 min). $^{1}$H-NMR (400 MHz, CDCl₃) δ 9.08 (d, Ar-H, $^3$J = 1.6 Hz, 1H), 8.74 (d, Ar-H, $^3$J = 1.8 Hz, 1H), 4.29 (m, CH₂-CH-CH₃, 1H), 4.11 (dd, CH₂-CH-CH₃, $^2$J = 8.0 Hz, 1H), 4.05 (m, CH₂-N-CH₂, 4H), 3.50 (t, CH₂-CH-CH₃, $^2$J = 8.0 Hz, 1H), 1.85 (m, CH₂-C-CH₂, 4H), 1.31 (d, CH₃, $^3$J = 6.1 Hz, 3H) $^{13}$C-NMR (125 Hz, CDCl₃) δ 166.5, 161.8, 143.9, 134.1, 133.3, 129.6 (q, $^1$J_C,F = 35.1 Hz), 126.6, 125.9, 122.4 (q, $^1$J_C,F = 273.1 Hz), 106.3, 72.5, 70.9, 44.6, 36.4, 35.2, 18.3 HRMS m/z calc for C₁₇H₁₇F₃N₃O₅S [M+H⁺], 432.0837; found, 432.0829, calc for C₁₇H₁₆F₃N₃O₅SNa [M+Na⁺], 454.0656; found, 454.0646.

Morpholine-4-carboxamide (30a) 1.0 g (16.70 mmol) urea were dissolved in 20 ml morpholine and refluxed for 40 h, until release of ammonia stopped. The amine was removed under reduced pressure and the resulting oily residue recrystallized from hexane:chloroform (approx. 1:2 (V/V)). Pale yellow crystals were collected and dried (1.64 g 76 %). $^{1}$H-NMR
(500 MHz, CDCl₃) δ 4.53 (bs, 2H, NH₂), 3.69 (m, 4H, CH₂-O-CH₂), 3.38 (m, 4H, CH₂-N-CH₂) MS m/z (EI) 130 (M).

2-(Morpholin-4-yl)-8-nitro-6-(trifluoromethyl)-4H-1,3-benzoxazin-4-one (30b) Synthesis of 2-chloro-3-nitro-5-(trifluoromethyl)benzoylchloride according to general procedure A from 250 mg (0.93 mmol) 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid. 151 mg (1.16 mmol) morpholine-4-carboxamide (30a) and 158 μl (0.93 mmol) DIPEA were dissolved in 30 ml toluene and heated to 70 °C. 2-chloro-3-nitro-5-(trifluoromethyl)benzoylchloride was dissolved in 4 ml toluene and added dropwise. Upon complete addition, the mixture was refluxed for 2.5 h. After cooling, the solvent was removed under reduced pressure and the crude product purified by flash chromatography (eluent TBME). The title compound appears as a pale-yellow solid (111 mg 35 %, HPLC purity 99.3 %, tₚ = 11.1 min). ¹H-NMR (400 MHz, CDCl₃) δ 8.71 (d, 1H, Ar-H, 4J = 2.3 Hz), 8.59 (d, 1H, Ar-H, 4J = 2.3 Hz), 3.93 (m, 4H, CH₂-N-CH₂), 3.83 (m, 4H, CH₂-O-CH₂) ¹³C-NMR (100 MHz, CDCl₃) δ 162.9, 155.4, 148.5, 136.3, 131.1 (q, JCF = 3.4 Hz), 127.8 (q, JCF = 35.9 Hz), 127.1 (q, JCF = 3.4 Hz), 122.1 (q, JCF = 273.1 Hz), 120.3, 66.2, 66.1, 45.4, 45.0 HRMS m/z calc for C₁₃H₁₁F₃N₃O₅ [M+H⁺], 346.0650; found, 346.0649, calc for C₁₃H₁₀F₃N₃O₅Na [M+Na⁺], 368.0466; found, 368.0467.

1,2,3,4-Tetrahydroisoquinoline-2-carboxamide (31a) Tetrahydroisoquinoline (1.000 g, 7.51 mmol, 2 eq.) and urea (0.225 g, 3.76 mmol, 1 eq.) are heated in a flask for 42 h at 120 °C (clear brown colouration). The crude product is worked up by recrystallisation from chloroform/hexane (1:1). The title compound appears as a brownish solid (479 mg 72 %). ¹H-NMR (400 MHz, DMSO-d₆) δ 7.13 (m, Ar-H, 4H), 6.00 (bs, NH₂, 2H), 4.45 (s, N-CH₂, 2H), 3.50 (t, N-CH₂, J = 5.9 Hz, 2H), 2.74 (t, CH₂, J = 5.9 Hz, 2H) MS m/z (APCI) 177.1 [M+H⁺].

8-Nitro-2-(1,2,3,4-tetrahydroisoquinolin-2-yl)-6-(trifluoromethyl)-4H-1,3-benzoxazin-4-one (31b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-
nitro-5-(trifluoromethyl)benzoic acid (0.337 g, 1.25 mmol, 1.25 eq.) by general procedure A. 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is dissolved in toluene (5 ml) and slowly added to a solution of 1,2,3,4-tetrahydroisoquinoline-2-carboxamide (31a) (0.200 g, 1.13 mmol, 1.0 eq.) and DIPEA (0.438 ml, 3.39 mmol, 3 eq.) in toluene at 70 °C. After 1 h reflux, the reaction is cooled overnight and the solvent was removed under vacuum. Purification is carried out by flash chromatography with the solvent chloroform. Subsequently, the reaction product is recrystallised from EtAc/hexane (1:1). The title compound appears as a pale yellowish solid (227 mg 58 %, HPLC purity 98.1 %, tR = 14.3 min). 1H-NMR (400 MHz, CDCl3) δ 8.74 (s, Ar-H, 1H), 8.61 (s, Ar-H, 1H), 7.23 (m, Ar-H, 4H), 5.03 (d, CH2, J = 8.0 Hz, 2H), 4.13 (m, CH2, 2H), 3.80 (s, CH3, 3H), 3.06 (m, CH2, 2H) 13C-NMR (100 MHz, CDCl3) δ 163.1, 162.9, 155.6, 155.5, 148.6, 136.3, 133.8, 133.4, 131.3, 131.1 (q, JCF = 3.8 Hz), 130.9, 128.7, 128.4, 127.6, 127.5 (q, JCF = 36.2 Hz), 127.4, 127.1, 127.0 (q, JCF = 3.8 Hz), 126.9, 126.5, 126.4, 122.2 (q, JCF = 272.8 Hz), 120.3, 47.1, 46.4, 43.6, 42.8, 28.6, 28.0 HRMS m/z calc for C18H13F3N3O4 [M+H+] = 392.0854; found, 392.0858.

Nitro-2-(1,2,3,4-tetrahydroisoquinolin-2-yl)-6-(trifluoromethyl)-1,4-dihydrochinazolin-4-one (32a) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.098 g, 0.36 mmol, 1.1 eq.) by general procedure A. 32a is synthesised by general procedure C, using 1,2,3,4-tetrahydroisoquinoline-2-guanidine hydroiodide (0.100 g, 0.33 mmol, 1.0 eq.). For a complete reaction, diazabicycloundecene (DBU) (0.039 g, 0.27 mmol, 0.82 eq.) is added as auxiliary base and the reaction is boiled for 3 h in toluene under reflux. The crude product is purified by flash chromatography (chloroform/MeOH 99:1). The title compound appears as a yellow solid (10 mg 8 %, HPLC purity 100.0 %, tR = 15.3 min). 1H-NMR (400 MHz, CDCl3) δ 8.55 (s, Ar-H, 1H), 8.29 (s, Ar-H, 1H), 7.26 (m, Ar-H, 4H), 4.99 (bs, N-CH2, 2H), 4.07 (t, N-CH2, J = 5.7 Hz, 2H), 3.08 (t, CH2, J = 5.7 Hz,
2H) $^{13}$C-NMR (100 MHz, CDCl$_3$) δ 163.4, 151.3, 134.2, 131.7 (q, $^3J_{CF} = 3.1$ Hz), 128.7, 128.4, 127.6, 127.0, 126.6 (q, $^3J_{CF} = 3.8$ Hz), 126.4, 123.4, 122.9 (q, $^2J_{CF} = 271.6$ Hz), 122.8, 118.6, 46.6, 42.9, 28.5 HRMS m/z calc for C$_{18}$H$_{14}$F$_3$N$_4$O$_3$ [M+H$^+$], 391.1013; found, 391.1002, calc for C$_{18}$H$_{13}$F$_3$N$_4$O$_3$Na [M+Na$^+$], 413.0833; found, 413.0821.

4-(Cyclohexylmethyl)piperazine-1-carboxamide (33a) Cyclohexylmethylpiperazine (0.500 g, 2.73, 2 eq.) and urea (0.082 g, 1.37 mmol, 1 eq.) are added to a flask and heated to 120 °C for 42 h (clear brown colouration). The crude product was purified up by flash chromatography with the eluent chloroform/MeOH/ammonia 36 % (95:5:1). The title compound appears as a brownish solid (308 mg 100 %).

$^1$H-NMR (400 MHz, CDCl$_3$) δ 4.55 (m, N$_2$H, 2H), 3.37 (m, C$_2$H$_2$-N-C$_2$H$_2$, 4H), 2.36 (m, C$_2$H$_2$-N-C$_2$H$_2$, 4H), 2.11 (d, N-C$_2$H$_2$-CH, $^3J = 7.0$ Hz, 2H), 1.70 (m, cyclohexyl, 5H), 1.47 (m, N-CH$_2$-C$_2$H, 1H), 1.18 (m, cyclohexyl, 3H), 0.86 (m, cyclohexyl, 2H)

2-[4-(Cyclohexylmethyl)piperazin-1-yl]-8-nitro-6-(trifluoromethyl)-4H-1,3-benoxazin-4-one (33b) 2-Chloro-3-nitro-5-(trifluoromethyl)benzoic acid chloride is prepared from 2-chloro-3-nitro-5-(trifluoromethyl)benzoic acid (0.266 g, 0.98 mmol, 1.0 eq.) by general procedure A. The carboxylic acid chloride is dissolved in toluene (5 ml) and slowly added to a solution of 4-(cyclohexylmethyl)piperazine-1-carboxamide (33a) (0.200 g, 0.89 mmol, 0.9 eq.) and DIPEA (0.499 ml, 2.94 mmol, 3 eq.) in toluene at 70 °C. After refluxing for 1 h, the reaction cooled overnight. The purification was carried out column chromatographically with the solvent medium chloroform/MeOH (98:2). The title compound appears as a brownish solid (162 mg 41 %, HPLC purity 95.8 %, $t_R = 11.3$ min). $^1$H-NMR (400 MHz, CDCl$_3$) δ 8.58 (d, Ar-H, $^4J = 2.3$, 1H), 8.45 (d, Ar-H, $^4J = 2.2$, 1H), 3.81 (m, CH$_2$-N-CH$_2$, 4H), 2.41 (m, CH$_2$-N-CH$_2$, 4H), 2.06 (m, N-CH$_2$-CH, 2H), 1.60 (m, cyclohexyl, 5H), 1.38 (m, N-CH$_2$-CH, 1H) 1.08 (m, cyclohexyl, 3H), 0.75 (m, cyclohexyl, 2H) $^{13}$C-NMR (125 MHz, CDCl$_3$) δ 163.0, 155.2, 148.6, 136.2, 131.0 (q, $^3J_{CF} = 3.8$ Hz), 127.5 (q, $^2J_{CF} = 35.3$ Hz), 127.0 (q, $^3J_{CF} = 3.8$ Hz), 122.1 (q, $^1J_{CF} = 273.3$ Hz), 118.6.
65.1, 53.0, 52.5, 45.2, 44.7, 34.9, 31.7, 26.6, 26.0 HRMS m/z calc for C_{20}H_{23}F_{3}N_{4}O_{4} [M+H^+],
441.1745; found, 441.1740
Figure S1A $^1$H-NMR (400 MHz) of 1c in CDCl$_3$.

Figure S1B $^{13}$C-NMR (100 MHz) of 1c in CDCl$_3$. 
Figure S1C HPLC chromatogram of 1c.
Figure S2A $^1$H-NMR (400 MHz) of 1e in CDCl$_3$.

Figure S2B $^{13}$C-NMR (100 MHz, CDCl$_3$) of 1e in CDCl$_3$. 
Figure S2C HPLC chromatogram of 1e.
Figure S3A $^1$H-NMR (500 MHz) of 2a in CDCl$_3$.

Figure S3B $^{13}$C-NMR (100 MHz) of 2a in CDCl$_3$. 

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Figure S3C HPLC chromatogram of 2a.
Figure S4A $^1$H-NMR (400 MHz) of 3c in CDCl$_3$.

Figure S4B $^{13}$C-NMR (100 MHz) of 3c in CDCl$_3$. 
Figure S4C HPLC chromatogram of 3c.
Figure S5A $^1$H-NMR (400 MHz) of 4c in CDCl$_3$.

Figure S5B $^{13}$C-NMR (100 MHz) of 4c in CDCl$_3$. 
Figure S5C HPLC chromatogram of 4c.
Figure S6A $^1$H-NMR (400 MHz) of 5c in CDCl$_3$.

Figure S6B $^{13}$C-NMR (100 MHz) of 5c in CDCl$_3$. 
Figure S6C HPLC chromatogram of 5c.
Figure S7A $^1$H-NMR (400 MHz) of 5d in CDCl$_3$.

Figure S7B $^{13}$C-NMR (100 MHz) of 5d in CDCl$_3$. 
Figure S7C HPLC chromatogram of 5d.
Figure S8A $^1$H-NMR (400 MHz) of 6d in CDCl$_3$.

Figure S8B $^{13}$C-NMR (100 MHz) of 6d in CDCl$_3$.
Figure S8C HPLC chromatogram of 6d.
Figure S9A $^1$H-NMR (400 MHz) of 7c in CDCl$_3$.

Figure S9B $^{13}$C-NMR (100 MHz) of 7c in CDCl$_3$. 
Figure S9C HPLC chromatogram of 7c.
Figure S10A $^1$H-NMR (400 MHz) of 8c in CDCl$_3$.

Figure S10B $^{13}$C-NMR (100 MHz) of 8c in CDCl$_3$. 
Figure S10C HPLC chromatogram of 8c.
**Figure S11A** $^1$H-NMR (400 MHz) of 9a in CDCl$_3$.

**Figure S11B** $^{13}$C-NMR (100 MHz) of 9a in CDCl$_3$. 
Figure S11C HPLC chromatogram of 9a.
Figure S12A $^1$H-NMR (400 MHz) of 10c in CDCl$_3$.

Figure S13B $^{13}$C-NMR (100 MHz) of 10c in DMSO-d$_6$.
Figure S12C HPLC chromatogram of 10c.
Figure S13A $^1$H-NMR (400 MHz) of 11c in CDCl$_3$.

Figure S13B $^{13}$C-NMR (100 MHz) of 11c in CDCl$_3$. 
Figure S13C HPLC chromatogram of 11c.
Figure S14A $^1$H-NMR (400 MHz) of 12b in CDCl$_3$.

Figure S14B $^{13}$C-NMR (100 MHz) of 12b in CDCl$_3$. 

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Figure S14C HPLC chromatogram of 12b.
Figure S15A ¹H-NMR (400 MHz) of 13b in CDCl₃.

Figure S15B ¹³C-NMR (100 MHz) of 13b in CDCl₃.
Figure S15C HPLC chromatogram of 13b.
Figure S16A $^1$H-NMR (400 MHz) of 14a in CDCl$_3$.

Figure S16B $^{13}$C-NMR (100 MHz) of 14a in CDCl$_3$. 
Figure S16C HPLC chromatogram of 14a.
Figure S17A $^1$H-NMR (400 MHz) of 15b in CDCl$_3$.

Figure S17B $^{13}$C-NMR (100 MHz) of 15b in CDCl$_3$. 
Figure S17C HPLC chromatogram of 15b.
Figure S18A $^1$H-NMR (400 MHz) of 16c in CDCl$_3$.

Figure S18B $^{13}$C-NMR (100 MHz) of 16c in CDCl$_3$. 

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Figure S18C HPLC chromatogram of 16c.
Figure S19A $^1$H-NMR (400 MHz) of 16d in CDCl$_3$.

Figure S19B $^{13}$C-NMR (125 MHz) of 16d in Acetone $d_6$. 

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Figure S19C HPLC chromatogram of 16d.
Figure S20A $^1$H-NMR (400 MHz) of 17b in CD$_3$OD.

Figure S20B $^{13}$C-NMR (100 MHz) of 17b in CD$_3$OD.
Figure S20C HPLC trace of 17b.
Figure S21A $^1$H-NMR (400 MHz) of 18b in CDCl$_3$.

Figure S21B $^{13}$C-NMR (100 MHz) of 18b in CDCl$_3$. 
Figure S21C HPLC chromatogram of 18b.
Figure S22A $^1$H-NMR (400 MHz) of 19b in CDCl$_3$.

Figure S22B $^{13}$C-NMR (100 MHz) of 19b in CDCl$_3$. 
Figure S22C HPLC chromatogram of 19b.
Figure S23A $^{1}$H-NMR (400 MHz) of 20b in CDCl$_3$.

Figure S23B $^{13}$C-NMR (100 MHz) of 20b in CDCl$_3$. 
Figure S23C HPLC chromatogram of 20b.
Figure S24A $^1$H-NMR (400 MHz) of 21b in CDCl$_3$.

Figure S24B $^{13}$C-NMR (100 MHz) of 21b in CDCl$_3$. 
Figure S24C HPLC chromatogram of 21b.
Figure S25A $^1$H-NMR (400 MHz) of $22b$ in CDCl$_3$.

Figure S25B $^{13}$C-NMR (100 MHz) of $22b$ in CDCl$_3$. 
Figure S25C HPLC chromatogram of 22b.
Figure S26A $^1$H-NMR (400 MHz) of 23b in CDCl$_3$.

Figure S26B $^{13}$C-NMR (100 MHz) of 23b in CDCl$_3$. 
Figure S26C HPLC trace of 23b.
Figure S27A $^1$H-NMR (400 MHz) of 24b in CDCl$_3$.

Figure S27B $^{13}$C-NMR (100 MHz) of 24b in CDCl$_3$. 
Figure S27C HPLC chromatogram of 24b.
Figure S28A $^1$H-NMR (400 MHz) of 26b in CDCl$_3$.

Figure S28B $^{13}$C-NMR (100 MHz) of 26b in CDCl$_3$. 
Figure S28C HPLC chromatogram of 25b.
Figure S29A $^1$H-NMR (400 MHz) of 26b in CDCl$_3$.

Figure S29B $^{13}$C-APT-NMR (100 MHz) of 26b in CDCl$_3$. 
Figure S29C HPLC chromatogram of 26b.
Figure S30A $^1$H-NMR (400 MHz) of 26c in CDCl$_3$.

Figure S30B $^{13}$C-APT-NMR (100 MHz) of 26c in CDCl$_3$. 
Figure S30C HPLC chromatogram of 26c.
Figure S31A $^1$H-NMR (400 MHz) of 26d in CDCl$_3$.

Figure S31B $^{13}$C-APT-NMR (100 MHz) of 26d in CDCl$_3$. 
Figure S31C HPLC trace of 26d.
**Figure S32A** $^1$H-NMR (400 MHz) of $27b$ in CDCl$_3$.

**Figure S32B** $^{13}$C-APT-NMR (125 MHz) of $27b$ in CDCl$_3$. 
Figure S32C HPLC chromatogram of 27b.
Figure S33A $^1$H-NMR (400 MHz) of *macozinone* in CDCl$_3$.

Figure S33B $^{13}$C-NMR (100 MHz) of *macozinone* in CDCl$_3$. 
Figure S33C HPLC trace of macozinone.
**Figure S34A** $^1$H-NMR (400 MHz) of **BTZ043** in CDCl$_3$.

**Figure S34B** $^{13}$C-NMR (100 MHz) of **BTZ043** in CDCl$_3$. 

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Figure S34C HPLC trace of BTZ043.

Sample Information
- Sample Name
- Tray# : 1
- Vial# : 30
- Injection Volume : 5 µL
- Data File : BTZ043.lcd
- Method File : MSP5-SS_30min_1.0.lcm
- Batch File : Batch170521_2.lcb
- Report Format File : Reportformat1.lsr
- Date Acquired : 18.05.2021 08:40:24
- Date Processed : 18.05.2021 09:19:28

| Peak# | Ret. Time | Area   | Height  | Area%  |
|-------|-----------|--------|---------|--------|
| 1     | 14.187    | 7587509| 652404  | 100.000|
| Total |           | 7587509| 652404  | 100.000|
Figure S35A $^1$H-NMR (400 MHz) of 30b in CDCl$_3$.

Figure S35B $^{13}$C-NMR (100 MHz) of 30b in CDCl$_3$. 
Figure S35C HPLC chromatogram of 30b.
Figure S36A $^1$H-NMR (400 MHz) of 31b in CDCl$_3$.

Figure S36B $^{13}$C-NMR (100 MHz) of 31b in CDCl$_3$. 
Figure S36C HPLC trace of 31b.
Figure S37A $^1$H-NMR (400 MHz) of 32a in CDCl$_3$.

Figure S37B $^{13}$C-NMR (100 MHz) of 32a in CDCl$_3$. 
Figure S37C HPLC chromatogram of 32a.
Figure S38A $^1$H-NMR (400 MHz) of 33b in CDCl$_3$.

Figure S38B $^{13}$C-NMR (125 MHz) of 33b in CDCl$_3$. 
Figure S38C HPLC chromatogram of 33b.
checkCIF/PLATON report

Structure factors have been supplied for datablock(s) ipds5968

THIS REPORT IS FOR GUIDANCE ONLY. IF USED AS PART OF A REVIEW PROCEDURE FOR PUBLICATION, IT SHOULD NOT REPLACE THE EXPERTISE OF AN EXPERIENCED CRYSTALLOGRAPHIC REFEREE.

No syntax errors found. CIF dictionary Interpreting this report

**Datablock: ipds5968**

Bond precision: C-C = 0.0023 Å  Wavelength=0.71073 Å

Cell:
- a=6.4887(3) Å
- b=9.4396(5) Å
- c=16.7854(8) Å
- alpha=80.347(4)°
- beta=82.041(4)°
- gamma=87.310(4)°

Temperature: 170 K

| Calculated       | Reported       |
|------------------|----------------|
| Volume           | 1003.50(9)     | 1003.50(9) |
| Space group      | P -1           | P -1       |
| Hall group       | -P 1           | -P 1       |
| Moiety formula   | C18 H19 F3 N4 O5 S | C18 H19 F3 N4 O5 S |
| Sum formula      | C18 H19 F3 N4 O5 S | C18 H19 F3 N4 O5 S |
| Mr               | 460.43         | 460.43     |
| Dx, g cm⁻³       | 1.524          | 1.524      |
| Z                | 2              | 2          |
| Mu (mm⁻¹)        | 0.228          | 0.228      |
| F000             | 476.0          | 476.0      |
| F000’            | 476.54         |            |
| h,k,lmax         | 8,12,23        | 8,12,23    |
| Nref             | 5428           | 5359       |
| Tmin, Tmax       | 0.909,0.934    | 0.745,1.328|
| Tmin’            | 0.894          |            |

Correction method= # Reported T Limits: Tmin=0.745 Tmax=1.328
AbsCorr = MULTI-SCAN

| Data completeness | 0.987 |
| Theta(max)       | 29.201|

R(reflections)= 0.0476( 4802)  wR2(reflections)= 0.1311( 5359)

S = 1.040  Npar= 293

The following ALERTS were generated. Each ALERT has the format test-name_ALERT_alert-type_alert-level.
Click on the hyperlinks for more details of the test.
| Alert level C |  |
|---------------|---|
| PLAT906_ALERT_3_C | Large K Value in the Analysis of Variance ...... 2.086 Check |
| PLAT911_ALERT_3_C | Missing FCF Refl Between Thmin & STh/L= 0.600 29 Report |
| PLAT913_ALERT_3_C | Missing # of Very Strong Reflections in FCF .... 9 Note |

| Alert level G |  |
|---------------|---|
| PLAT002_ALERT_2_G | Number of Distance or Angle Restraints on AtSite 8 Note |
| PLAT003_ALERT_2_G | Number of Uiso or Uij Restained non-H Atoms ... 6 Report |
| PLAT151_ALERT_1_G | The s.u.’s on the Cell Angles are Equal ..(Note) 0.004 Degree |
| PLAT171_ALERT_4_G | The CIF-Embedded .res File Contains EADP Records 3 Report |
| PLAT176_ALERT_4_G | The CIF-Embedded .res File Contains SADI Records 3 Report |
| PLAT186_ALERT_4_G | The CIF-Embedded .res File Contains ISOR Records 1 Report |
| PLAT230_ALERT_2_G | Hirshfeld Test Diff for F1' --C9 . 20.5 s.u. |
| PLAT230_ALERT_2_G | Hirshfeld Test Diff for F2' --C9 . 5.2 s.u. |
| PLAT230_ALERT_2_G | Hirshfeld Test Diff for F3' --C9 . 6.7 s.u. |
| PLAT242_ALERT_2_G | Low ‘MainMol’ Ueq as Compared to Neighbors of C9 Check |
| PLAT301_ALERT_3_G | Main Residue Disorder ..............(Resd 1 ) 10% Note |
| PLAT860_ALERT_3_G | Number of Least-Squares Restraints ............. 81 Note |
| PLAT910_ALERT_3_G | Missing # of FCF Reflection(s) Below Theta(Min). 1 Note |
| PLAT912_ALERT_4_G | Missing # of FCF Reflections Above STh/L= 0.600 40 Note |
| PLAT933_ALERT_2_G | Number of OMIT Records in Embedded .res File ... 2 Note |
| PLAT994_ALERT_3_G | Average HKL Measurement Multiplicity ........... 2.7 Low |
| PLAT978_ALERT_2_G | Number C-C Bonds with Positive Residual Density. 11 Info |

0 ALERT level A = Most likely a serious problem - resolve or explain
0 ALERT level B = A potentially serious problem, consider carefully
3 ALERT level C = Check. Ensure it is not caused by an omission or oversight
17 ALERT level G = General information/check it is not something unexpected

1 ALERT type 1 CIF construction/syntax error, inconsistent or missing data
8 ALERT type 2 Indicator that the structure model may be wrong or deficient
7 ALERT type 3 Indicator that the structure quality may be low
4 ALERT type 4 Improvement, methodology, query or suggestion
0 ALERT type 5 Informative message, check
It is advisable to attempt to resolve as many as possible of the alerts in all categories. Often the minor alerts point to easily fixed oversights, errors and omissions in your CIF or refinement strategy, so attention to these fine details can be worthwhile. In order to resolve some of the more serious problems it may be necessary to carry out additional measurements or structure refinements. However, the purpose of your study may justify the reported deviations and the more serious of these should normally be commented upon in the discussion or experimental section of a paper or in the "special_details" fields of the CIF. checkCIF was carefully designed to identify outliers and unusual parameters, but every test has its limitations and alerts that are not important in a particular case may appear. Conversely, the absence of alerts does not guarantee there are no aspects of the results needing attention. It is up to the individual to critically assess their own results and, if necessary, seek expert advice.

**Publication of your CIF in IUCr journals**

A basic structural check has been run on your CIF. These basic checks will be run on all CIFs submitted for publication in IUCr journals (Acta Crystallographica, Journal of Applied Crystallography, Journal of Synchrotron Radiation); however, if you intend to submit to Acta Crystallographica Section C or E or IUCrData, you should make sure that full publication checks are run on the final version of your CIF prior to submission.

**Publication of your CIF in other journals**

Please refer to the Notes for Authors of the relevant journal for any special instructions relating to CIF submission.

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