Synthesis and biological evaluation of 5-substituted O^4-alkylpyrimidines as CDK2 inhibitors

Francesco Marchetti,[a] Celine Cano,[a] Nicola J. Curtin,[b] Bernard T. Golding,[c] Roger J. Griffin,[a] Karen Haggerty,[a] David R. Newell,[b] Rachel J. Parsons,[a] Sara L. Payne,[a] Lan Z. Wang[b] and Ian R. Hardcastle[a]*

Supporting information

EXPERIMENTAL

Reagents were purchased from fine chemicals vendors, and used as received unless otherwise stated. Solvents were purified and stored according to standard procedures. Petrol refers to that fraction in the boiling range 40-60 °C. Melting points were obtained on a Stuart Scientific SMP3 apparatus and are uncorrected. Thin layer chromatography was performed using silica gel plates (Kieselgel $60F_{254}$; 0.2 mm), and visualized with UV light or potassium permanganate. Chromatography was conducted under medium pressure in glass columns or using a Biotage SP4 instrument in prepacked columns (FLASH+ Silica columns (40-63 µm, 60 Å), NH FLASH+ cartridges (40-75 µm, 100 Å) and C18 FLASH+ cartridges (40-63 µm, 90 Å)). Semi-preparative HPLC was conducted using using a Varian Prostar instrument equipped with a Waters XTerra column (RP_{18} OBD 10 µm, 150 x 30 mm) or Phenomenex Synergi 4u Fusion-RP (80Å, 250 x 21.2 mm) as indicated, monitoring by UV at λ = 270 nm. Proton (^{1}H) and carbon (^{13}C) nuclear magnetic resonance (NMR) spectra were recorded on a Bruker Spectrospin AC 300E (^{1}H at 300 MHz, ^{13}C at 75 MHz) or a Jeol JNM-LA500 spectrometer (^{1}H at 500 MHz, ^{13}C at 125 MHz) employing the solvent as internal standard. Coupling constants *J* values are given in Hz. NH signals appeared as broad singlets (br s) exchangeable with D_2O. Mass spectra were determined on a Micromass Autospec M spectrometer in electron impact (EI) mode. Liquid Chromatography-Mass Spectrometry (LCMS) was carried out on a Micromass Platform instrument operating in positive and negative ion electrospray mode, employing either a Waters Symmetry (30 x 4.6 mm; C18; 5 min run) or Waters Atlantis (50 x 4.6 mm; C18; 12 min run) column with 0.05% aqueous formic acid and acetonitrile (5 to 95% organic). Elemental combustion analyses were either recorded on Carlo-Erba instrument 1106 analyser, in-house, or were performed either by Medac Ltd (Brunel Science Centre, Egham, Surrey, TW20 0JZ), or by the School of Pharmacy at London University (29-39 Brunswick Square, London, WC1N 1AX). Accurate mass analyses were measured using a Finnigan MAR 95 XP or a Finnigan MAR 900 XLT at Swansea EPSRC National Mass Spectrometry Service Centre (EPSRC, Chemistry Department, University of Wales Swansea, Wales, SA2 8PP). Final compounds were found to be >97% purity by LCMS and NMR analysis or gave satisfactory CNH analysis.

General procedure A

Sodium (1.5 eq.) was added to the appropriate alcohol (20 mL) and the resulting mixture was stirred and heated for 2 h, then 4-chloro-2,6-diaminopyrimidine (1 eq.) was added and stirring continued 5h with heating to the specified temperature. After cooling, the mixture was neutralised with glacial acetic acid and concentrated *in vacuo* to yield a yellow oil, which was diluted with water (25 mL) and extracted with EtOAc (4 x 25 mL). The combined organic layers were dried ($MgSO_4$) and concentrated *in vacuo* giving the desired product.

6-Ethoxypyrimidine-2,4-diamine (3b)

General procedure A: Sodium (0.92 g, 40 mmol), ethanol (40 mL, 696 mmol), 4-chloro-2,6-diaminopyrimidine (1.15 g, 8.0 mmol), at 80 °C. Chromatography (silica; 5% MeOH, DCM) gave **3b** as a light brown powder (0.35 g, 29%) mp 167-169 °C. UV λ_{max} (EtOH): 206 nm; IR υ_{max}/ cm^{-1}: 3329, 2985, 1661, 1580; δ_H (300 MHz, d_6-DMSO) = 1.21 (3H, t, *J* = 7.1 Hz, C**H**$_3$), 4.12 (2H, q, *J* = 7.1 Hz, C**H**$_2$), 0.88-0.93 (1H, m, C**H**), 5.01 (1H, s, H-5), 5.83 (2H, s, NH_2 ex), 5.98 (2H, s, NH_2 ex); δ_C (75 MHz, d_6-DMSO) δ = 15.1, 60.5, 76.4, 163.3, 166.3, 170.3; MS (ESI+) *m*/*z* 155 $[M+H]^+$.

6-Cyclopropylmethoxypyrimidine-2,4-diamine (3c)

General procedure A: Sodium (0.70 g, 30 mmol, 1.5 eq.), cyclopropylmethanol (20 mL, 247 mmol), 4-chloro-2,6-diaminopyrimidine (0.86 g, 5.9 mmol), at 140 °C. Gave **3c** as a yellow oil (4.878 g, 96.7%). UV λ_{max} (EtOH): 266 nm**;** IR υ_{max}/ cm^{-1}: 3329, 2947, 1562; δ_H (300 MHz, d_6-DMSO): 0.01 (2H, m, C**H**$_2$), 0.24-0.30 (2H, m, C**H**$_2$), 0.88-0.93 (1H, m, C**H**), 3.68 (2H, d, *J* = 7.1 Hz, C**H**$_2$O), 4.79 (1H, s, H-5), 5.61 (2H, s, NH_2 ex), 5.75 (2H, s, NH_2 ex) ; MS (ESI+) *m*/*z* 181 $[M+H]^+$;

6-*sec*-Butoxypyrimidine-2,4-diamine (3d)

General procedure A: Sodium (0.95 g, 41.8 mmol), *sec*-2-butanol (35.0 mL, 382 mmol), 4-chloro-2,6-diaminopyrimidine (4.00 g, 27.7 mmol), at 100 °C. Gave **3d** as a yellow oil (4.878 g, 96.7%). UV λ_{max} (EtOH): 266, 235, 214 nm**;** IR υ_{max}/ cm^{-1}: 3326, 3184, 2971, 2934, 1560, 1441, 1200; δ_H (300 MHz, d_6-DMSO): 0.84 (3H, t, *J* = 7.5 Hz, $CH_3CHORCH_2C\mathbf{H}_3$), 1.14 (3H, d, *J* = 6.2 Hz, $C\mathbf{H}_3CHORCH_2CH_3$), 1.44-1.60 (2H, m, $CH_3CHORC\mathbf{H}_2CH_3$), 4.93 (H, m, $CH_3C\mathbf{H}ORCH_2CH_3$), 5.01 (1H, s, H-5), 5.88 (2H, s, NH_2 ex), 5.99 (2H, s, NH_2 ex); δ_C (75 MHz, d_6-DMSO): 9.6, 19.5, 28.7, 70.9, 76.8, 163.0, 166.0, 169.9 ; MS (ESI+) *m*/*z* 183 $[M+H]^+$; HRMS (ESI+) *m*/*z*: Calc. for $C_8H_{14}N_4O$: 183.1240 $[M+H]^+$. Found 183.1242 $[M+H]^+$.

2,6-Diamino-4-heptoxypyrimidine (3e).

A mixture of heptan-1-ol (1.1 ml, 7.8 mmol) and sodium hydride (0.10 g, 4.3 mmol) in DMSO (4 ml) was stirred at rt for 30 min. 4-Chloro-2,6-diaminopyrimidine (0.21 g, 1.4 mmol) was added, and the mixture was heated at 85 °C for 4 days. The reaction mixture was allowed to cool to rt and then neutralised with glacial acetic acid; the solvents were removed *in vacuo*. Chromatography (silica; 10% MeOH, DCM) gave **3e** as a cream solid (0.14 g, 43%) m.p. 90.8–92.6 ºC. UV (EtOH): λ_{max} = 240 nm, ε = 8.21 $dm^3\ g^{-1}\ cm^{-1}$; IR ν_{max}/ 3211, 3118 cm^{-1}; δ_H (200 MHz, d^6-DMSO) δ 0.90 (3H, t, *J* = 6.6 Hz, CH_3), 1.31 (8H, br s, 4 x CH_2), 1.63 (2H, m, $OCH_2\mathbf{CH_2}$), 4.09 (2H, t, *J* = 6.5 Hz, OCH_2), 5.04 (1H, s, H5), 5.87 (2H, br s, D_2O exch, NH_2), 6.01 (2H, br s, D_2O exch, NH_2). MS $(ESI)^+$ *m/z* = 225.13, $[M+H]^+$. $C_{11}H_{20}N_4O.CH_3CO_2H$ requires: C, 54.91; H, 8.51; N, 19.70; found: C, 53.36; H, 8.52; N, 18.83%

General Procedure B

A mixture of 5-bromo-6-cyclohexylmethoxypyrimidine-2,4-diamine (**5**) (0.6 g, 1.99 mmol, 1.0 eq.), the appropriate aryl or heteroaryl boronic acid (2.39 mmol, 1.2 eq.), tetrakis(triphenylphosphine)palladium(0) (0.115 g, 0.1 mmol, 5% mol eq.), K_2CO_3 solution (4 M, 2 mL, 8 mmol, 4.0 eq.), and 1,2-dimethoxyethane (10 mL) was heated under microwave irradiation at 170 °C, for 1-2 h. The resulting mixture was filtered through Celite, washed successively with MeOH (4 x 30 mL) and DCM (4 x 30 mL). The combined fractions were concentrated *in vacuo*. To remove further palladium residues, a thiol-functionalized resin PL-Thiol SPE Tube (Stratospheres^{TM} SPE) was employed. The SPE tube was preconditioned with methanol, then the compound sample, previously disolved in the smallest amount possible of methanol, was loaded and allowed to pass through the SPE media under gravity and the filtrate concentrated *in vacuo.* The residues were disolved in water (30 mL) and extracted with EtOAc (3 x 30 mL). The combined organic fractions were dried (Na_2SO_4) and concentrated *in vacuo*. Chromatography (silica; EtOAc, petrol) followed by HPLC, Waters XTerra column, gave the desired product **7a-c** as a white solid.

6-Cyclohexylmethoxy-5-(furan-2-yl)pyrimidine-2,4-diamine (7b)

General procedure B: 2-furan boronic acid (0.27 g, 2.39 mmol, 1.2 eq.). Chromatography and HPLC gave **7b**, Waters XTerra column, as an off-white solid (0.083 g, 15%). m.p. 91-94 °C. UV λ_{max} (EtOH) 286 and 210 nm. IR ν_{max} 3329, 3109, 2920-2984, 1565, 1542, 1426, 1208, 1074 cm^{-1}; δ_H (300 MHz, d_3-MeCN): 1.02-1.84 (11H, m, C_6H_{11}), 4.09 (2H, d, *J* = 6.0 Hz, OCH_2), 4.98 (2H, s, NH_2 ex), 5.89 (2H, s, NH_2 ex), 6.50 (1H, m, ArH), 6.65 (1H, d, *J* = 3.8 Hz, ArH), 7.43 (1H, d, *J* = 1.8 Hz, ArH) ; ^{13}C-NMR (75 MHz, $CDCl_3$): 26.1, 26.8, 30.3, 37.8, 72.5, 85.6, 108.8, 111.6, 140.0, 148.4, 159.3, 160.3, 167.6 . MS (ESI+) *m*/*z* 289.16 $[M+H]^+$; HRMS (ESI+) *m/z*: Calc. for $C_{15}H_{20}N_4O_2$: 289.1659 $[M+H]^+$. Found 289.1662 $[M+H]^+$. $C_{15}H_{20}N_4O_2.0.2\ CHCl_3$: requires C, 59.51; H, 6.71; N, 18.24%; found C, 59.18; H, 6.47, N, 17.96%.

6-Cyclohexylmethoxy-5-(thiophen-2-yl)pyrimidine-2,4-diamine (7c)

General procedure B: 2-thiophene boronic acid (0.27 g, 2.39 mmol, 1.2 eq.). Chromatography and HPLC, Waters XTerra column, gave the **7c** as an off-white solid (0.114 g, 19%), m.p.: 84-86 °C. UV λ_{max} (EtOH): 378 and 244 nm. IR ν_{max} 3392, 3196, 2920-2840, 1540, 1444, 1418, 1337, 1143 cm^{-1}; δ_H (300 MHz, $CDCl_3$): 0.81-1.65 (11H, m, C_6H_{11}), 3.93 (2H, d, *J* = 9.0 Hz, OCH_2), 5.03 (2H, s, NH_2 ex), 5.07 (2H, s, NH_2 ex), 6.92 (1H, br, ArH), 6.95 (1H, d, *J* = 4.5 Hz, ArH), 7.24 (1H, d, *J* = 4.5 Hz, ArH) ; ^{13}C-NMR (75 MHz, $CDCl_3$): 26.1, 26.9, 30.1, 37.8, 71.8, 86.9, 126.0, 127.0, 127.6, 135.1, 161.8, 163.6, 168.2; MS (ESI+) *m/z* 305.21 $[M+H]^+$. HRMS (ESI+) *m/z*: Calc. for $C_{15}H_{20}N_4OS$: 305.1431 $[M+H]^+$. Found 305.1430 $[M+H]^+$.

General procedure C

A mixture of 6-amino-2-(*n*-butylsulfanyl)-4(3H)-pyrimidinone (**15**) (1 eq.), the appropriate alcohol (1.5 eq.), and triphenylphosphine (1.5 eq.) in THF (anh) was stirred at 5 °C in an ice-bath and diethyl azodicarboxylate (1.5 eq.) was added dropwise. The resulting mixture was allowed to stir at rt for 72 h then concentrated *in vacuo* to yield a yellow oil, which was triturated with Et_2O in an ice-bath at 0 °C, giving a white precipitate which was removed by filtration. The filtrate was concentrated *in vacuo*, water (30 mL) was added and the mixture was extracted with EtOAc (3 x 30 mL). The combined organic layers were dried (Na_2SO_4) and concentrated *in vacuo*. Chromatography (silica; 20-80% EtOAc, petrol) and recrystallisation (MeOH) gave the desired product **16a-c**.

6-*sec*-Butoxy-2-(*n*-butylsulfanyl)pyrimidin-4-amine (16b)

General procedure C: 15 (0.5 g, 2.51 mmol), *sec*-2-butanol (2.15 mL, 29 mmol), triphenylphosphine (0.99 g, 3.77 mmol), THF (15.0 mL), diethyl azodicarboxylate (0.549 mL, 3.77 mmol) gave **16b** as a yellow oil (0.54 g, 85%). UV λ_{max} (EtOH): 224, 253 nm; IR υ_{max}/ cm^{-1}: 3469, 3352, 3181, 2964, 2931, 2872, 1620, 1573, 1545, 1343, 1231, 1035. δ_H (300 MHz, d_6-DMSO): 0.86 (3H, t, *J* = 7.3 Hz, C**H**$_3$$CH_2CH_2CH_2S$), 0.89 (3H, t, *J* = 7.4 Hz, $CH_3CHORCH_2$C**H**$_3$), 1.20 (3H, d, *J* = 6.2 Hz, C**H**$_3$$CHORCH_2CH_3$), 1.32-1.44 (2H, m, CH_3C**H**$_2$$CH_2CH_2S$), 1.51-1.66 (4H, m, CH_3CH_2C**H**$_2$$CH_2S$, CH_3CHORC**H**$_2CH_3$), 2.97 (2H, t, *J* = 7.4 Hz, $CH_3CH_2CH_2$C**H**$_2$S), 5.00 (1H, m, CH_3C**H**$ORCH_2CH_3$), 5.38 (1H, s, H-5), 6.55 (2H, s, NH_2 ex) ; δ_C (75 MHz, d_6-DMSO): 9.6, 13.5, 19.4, 21.6, 28.5, 29.6, 31.6, 72.4, 82.1, 165,2, 168,4, 169.2 ; MS (ESI+) *m/z* 256.13 $[M+H]^+$; HRMS (ESI+) *m/z*: Calc. for $C_{12}H_{21}N_3OS$: 256.1478 $[M+H]^+$. Found 256.1477 $[M+H]^+$.

2-(*n*-Butylsulfanyl)-6-ethoxypyrimidin-4-amine (16c)

General procedure C: 15 (4.5 g, 22.6 mmol), ethanol (2.15 mL, 33 mmol,), triphenylphosphine (8.88 g, 33.9 mmol), THF (150 mL), diethyl azodicarboxylate (5.34 mL, 34 mmol) gave **16c** as a white solid (3.6 g, 70%). mp 88-90 °C. UV λ_{max} (EtOH): 211, 223, 252 nm; IR υ_{max}/ cm^{-1}: 3378, 3303, 3158, 2958, 2932, 1636, 1573, 1543, 1373, 1266, 1009. δ_H (300 MHz, d_6-DMSO): 0.88 (3H, t, *J* = 7.3 Hz, C**H**$_3$$CH_2CH_2CH_2S$), 1.25 (3H, t, *J* = 7.1 Hz, OCH_2C**H**$_3$), 1.32-1.44 (2H, m, CH_3C**H**$_2$$CH_2CH_2S$), 1.55-1.66 (2H, m, CH_3CH_2C**H**$_2$$CH_2S$), 3.01 (2H, t, *J* = 7.3 Hz, $CH_3CH_2CH_2$C**H**$_2$S), 4.21 (3H, t, *J* = 7.1 Hz, OC**H**$_2CH_3$), 5.39 (1H, s, H-5), 6.65 (2H, s, NH_2 ex) ; δ_C (75 MHz, d_6-DMSO): 13.8, 14.8, 21.9, 29.7, 31.9, 63.0, 81.9, 165.5, 168.9, 169.6 ; MS (ESI+) *m/z* 228 $[M+H]^+$; HRMS (ESI+) *m/z*: Calc. for $C_{10}H_{17}N_3OS$: 228.1165 $[M+H]^+$. Found 228.1162 $[M+H]^+$.

General Procedure D

To a solution of the appropriate 2-(*n*-butylsulfanyl)-6-alkoxyoxypyrimidin-4-amine (**16a-c**) (1 eq.) in DCM under N_2, *m*-CPBA (2.5–3 eq.) was added slowly. The resulting mixture was stirred at rt for 24 h, then concentrated *in vacuo*. The residue was extracted with EtOAc (3 x 30 mL). The combined organic fractions were washed with aq. sodium sulfite (sat.; 30 mL), aq. sodium bicarbonate (sat.; 2 x 30 mL) and water (2 x 30 mL), dried (Na_2SO_4) and concentrated *in vacuo* to give a white solid. Chromatography (silica; 20-80% EtOAc, petrol) gave **17a-c** as a white solid.

6-*sec*-Butoxy-2-(*n*-butylsulfonyl)pyrimidin-4-amine (17b)

General procedure D: 16b (0.54 g, 2.11 mmol), DCM (15 mL), *m*-CPBA (1.46 g, 6.34 mmol, 3 eq.) gave **17b** as a yellow solid (0.3 g, 50%) m.p.: 74-77 °C.

UV λ_{max} (EtOH): 281, 244 nm; IR υ_{max}/ cm^{-1}: 3443, 3331, 3133, 2967, 2938, 2877, 1634, 1592, 1527, 1443, 1379, 1234; δ_H (300 MHz, $CDCl_3$): 0.89 (6H, m, C**H**$_3$CH$_2$CH$_2$CH$_2$SO$_2$, CH$_3$CHORCH$_2$C**H**$_3$), 1.25 (3H, d, *J* = 6.1 Hz, C**H**$_3$CHORCH$_2$CH$_3$), 1.37-1.49 (2H, m, CH$_3$C**H$_2$**CH$_2$CH$_2$SO$_2$), 1.53-1.69 (2H, m, CH$_3$CHORC**H$_2$**CH$_3$), 1.72-1.82 (2H, m, CH$_3$CH$_2$C**H$_2$**CH$_2$SO$_2$), 3.37 (2H, t, *J* = 7.4 Hz, CH$_3$CH$_2$CH$_2$C**H$_2$**SO$_2$), 4.98-5.08 (1H, m, CH$_3$C**H**ORCH$_2$CH$_3$), 5.78 (1H, s, H-5), 6.01 (2H, s, NH_2 ex) ; δ_C (75 MHz, $CDCl_3$): 9.3, 13.3, 19.2, 21.7, 24.2, 28.9, 50.9, 75.0, 89.2, 164.5, 165.6, 170.6 . MS (ESI+) *m*/*z* 288.25 $[M+H]^+$; HRMS (ESI+) *m/z*: Calc. for $C_{12}H_{21}N_3O_3S$: 288.1376 $[M+H]^+$. Found 288.1380 $[M+H]^+$.

2-(*n*-Butylsulfonyl)-6-ethoxypyrimidin-4-amine (17c)

General procedure D: 16c (2.90 g, 12.8 mmol, 1 eq.) DCM (50 mL), *m*-CPBA (7.34 g, 31.9 mol, 2.5 eq.) gave **17c** as a white solid (2.23 g, 67.4%) m.p. 136-139 °C. UV λ_{max} (EtOH): 215 and 270 nm; IR υ_{max} 3430, 3316, 3103, 2964, 2933, 2874, 1635, 1595, 1529, 1475, 1381, 1286 cm^{-1}; δ_H (300 MHz, d_6-DMSO 0.88 (3H, t, *J* = 7.3 Hz, C**H**$_3$CH$_2$CH$_2$CH$_2$SO$_2$), 1.29 (3H, t, *J* = 7.1 Hz, C**H**$_3$CH$_2$O), 1.34-1.46 (2H, m, CH$_3$C**H$_2$**CH$_2$CH$_2$SO$_2$), 1.61-1.70 (2H, m, CH$_3$CH$_2$C**H$_2$**CH$_2$SO$_2$), 3.43 (2H, t, *J* = 7.9 Hz, CH$_3$CH$_2$CH$_2$C**H$_2$**SO$_2$), 4.26 (2H, q, *J* = 7.1 Hz, CH$_3$C**H$_2$**O), 5.81 (1H, s, H-5), 7.38 (2H, s, NH_2 ex) ; δ_C (75 MHz, d_6-DMSO): 13.8, 14.7, 21.4, 24.3, 49.9, 62.9 , 87.3, 164.6, 166.5, 169.6 . MS (ESI+) *m*/*z* 260.14 $[M+H]^+$; HRMS (ESI+) *m/z*: Calc. for $C_{10}H_{17}N_3O_3S$: 260.1063 $[M+H]^+$. Found 260.1065 $[M+H]^+$.

General procedure E

A solution of the required pyrimidine (0.50 g, 2.62 mmol, 1 eq.) and the chosen aniline (5.76 mmol, 2.2 eq.) in trifluoroethanol (10 mL) with trifluoroacetic acid (1.0 mL, 13.1 mmol, 5 eq.) was stirred for 48h at rt or heated at reflux 24h as specified. The solvents were removed *in vacuo*, water (30 mL) was added and the pH was adjusted to neutral with saturated aq. sodium bicarbonate and the mixture was extracted with EtOAc (3 x 30 mL). The combined organic layers were washed with water (2 x 30 mL) and dried (Na_2SO_4), and the solvent was removed *in vacuo*. The compounds were purified as specified.

4-Amino-6-chloro-2-(4-methoxyphenylamino)-5-formylpyrimidine (21b)

General procedure E: 4-amino-2,6-dichloro-5-formylpyrimidine (**20**) (0.5 g, 2.62 mmol), p-anisidine (0.71 g, 5.76 mmol,), trifluoroacetic acid (0.48 mL, 6.51 mmol) and trifluoroethanol (10 mL). Chromatography (silica, 20-80% EtOAc, petrol) and recrystallisation (methanol), gave the **21b** as a dark yellow solid (0.61 g, 83.6%) m.p.: 185-186 °C. UV λ_{max} (EtOH): 225 and 337 nm. IR ν_{max}/ cm^{-1}: 3343, 3283, 3073, 2935, 1716, 1540, 1502, 1232. δ_H (300 MHz, $CDCl_3$): 3.74 (3H, s, OCH_3), 5.74 (1H, br, NH), 6.81 (2H, d, *J* = 9.1 Hz, ArH), 7.26 (1H, br, NH), 7.37 (2H, d, *J* = 9.1 Hz, ArH), 8.77 (1H, br, NH), 10.05 (1H, s, CHO) . δ_C (75 MHz, $CDCl_3$): 55.9, 103.5, 114.8, 123.4, 130.9, 157.4, 159.7, 164.3, 166.0, 188.7 . MS (ESI+) *m*/*z* 279.13 $[M+H]^+$. HRMS (ESI+) *m/z*: Calc. for $C_{12}H_{11}ClN_4O_2$: $[M+H]^+$ 279.0643. Found 279.0643. $C_{17}H_{11}ClN_4O_2$: requires C, 51.72; H, 3.98; N, 20.10%. Found C, 51.95; H, 3.95, N, 19.91%.

Neopentyl 4-(4-amino-6-chloro-5-formylpyrimidin-2-ylamino)benzoate (21c)

General procedure E: 4-amino-2,6-dichloro-5-formylpyrimidine (**20**) (0.84 g, 4.4 mmol), neopentyl 4-aminobenzoate[21] (1.00 g, 4.83 mmol), trifluoroacetic acid (1.7 mL, 22 mmol) and trifluoroethanol (30 mL). Chromatography (silica, 20-80% EtOAc, petrol) and recrystallisation (methanol), gave **21c** as a yellow solid (0.55 g, 35%) m.p.: 231-233 °C. UV λ_{max} (EtOH): 222 and 337 nm. IR ν_{max}/ cm^{-1}: 3314, 3078, 2954, 2847, 1689, 1656, 1575, 1535, 1358. δ_H (300 MHz, $CDCl_3$): 1.04 (9H, s, CH_3), 4.00 (2H, s, OCH_2), 5.87 (1H, br, NH), 7.52 (1H, br, NH), 7.69 (2H, d, *J* = 8.8 Hz, ArH), 8.05 (2H, d, *J* = 8.8 Hz, ArH), 8.92 (1H, br, NH), 10.17 (1H, s, CHO) . δ_C (75 MHz, $CDCl_3$): 26.9, 31.9, 74.5, 104.1, 119.8, 126.5, 131.0, 142.2, 159.3, 164.3, 166.1, 166.3, 188.9 . MS (ESI+) *m*/*z* 363.24 $[M+H]^+$. HRMS (ESI+) *m/z*: Calc. for

$C_{17}H_{19}ClN_4O_3$: 363. 1218 $[M+H]^+$. 363.1214 Found $[M+H]^+$. $C_{17}H_{19}ClN_4O$ requires C, 56.28; H, 5.28; N, 15.44%; found C, 56.51; H, 5.21, N, 15.62%.

4-(4-Amino-6-sec-butoxypyrimidin-2-ylamino)benzenesulfonamide (18b)

General procedure E: 6-*sec*-Butoxy-2-(*n*-butylsulfonyl)pyrimidin-4-amine (**17b**) (0.6 g, 2.09 mmol) and 4-aminobenzensulfonamide (0.72 g, 4.17 mmol) in TFE (6 mL) with TFA (0.8 mL, 10.4 mmol), heated at reflux for 24 h. Chromatography (silica; 30-95% EtOAc, petrol, 5% formic acid) and recrystalisation (MeOH) gave **18b** as a pale yellow solid (0.47 g, 67%) m.p. 111-113 °C. UV λ_{max} (EtOH): 271 nm. IR υ_{max}/ cm^{-1}: 3412, 3287, 3097, 2925-2844, 1641, 1557, 1437, 1367, 1192. δ_H (300 MHz, $CDCl_3$): 0.89 (3H, t, *J* = 7.4 Hz, $CH_3CHORCH_2C\mathbf{H}_3$), 1.25 (3H, d, *J* = 6.2 Hz, $C\mathbf{H}_3CHORCH_2CH_3$), 1.61-1.76 (2H, m, $CH_3CHORC\mathbf{H}_2CH_3$), 4.91-5.01 (1H, m, $CH_3C\mathbf{H}ORCH_2CH_3$), 4.57 (2H, br, NH_2 exchangeable with D_2O), 4.81 (2H, br, NH_2 exchangeable with D_2O), 5.32 (1H, s, H-5), 7.19 (1H, br, NH, exchangeable with D_2O), 7.68 (2H, d, *J* = 8.9 Hz, ArH), 7.76 (2H, d, *J* = 8.9 Hz, ArH) ; δ_C (75 MHz, $CDCl_3$): 9.7, 19.5, 29.1, 73.8, 83.2, 118.2, 127.8, 134.0, 144.4, 158.7, 164.9, 170.9 ; MS (ESI+) *m*/*z* 338.29 $[M+H]^+$. HRMS (ESI+) *m/z*: Calc. for $C_{14}H_{19}N_5O_3S$: 338.1281 $[M+H]^+$. Found 338.1283 $[M+H]^+$.

4-(4-Amino-6-ethoxypyrimidin-2-ylamino)benzensulfonamide (18c)

General procedure E: 2-(*n*-Butylsulfonyl)-6-ethoxypyrimidin-4-amine (**17c**) (0.6 g, 2.32 mmol) and 4-aminobenzensulfonamide (0.60 g, 3.47 mmol), TFE (8 mL), TFA (0.89 mL, 11.6 mmol), heated at reflux for 24 h. Chromatography (silica; 20-95% EtOAc, 5% formic acid, petrol) gave **18c** as a white solid (0.45 g, 63%) m.p.: 214-217 °C. UV λ_{max}(EtOH): 295 nm. IR υ_{max}/ cm^{-1}: 3435, 3317, 3106, 2913, 2856, 1616, 1553, 1422, 1375 (NH_2SO_2), 1195 (SO_2). δ_H (300 MHz, d_6-DMSO): 1.29 (3H, t, *J* = 7.1 Hz, $C\mathbf{H}_3CH_2O$), 4.24 (2H, q, *J* = 7.1 Hz, $CH_3C\mathbf{H}_2O$), 5.29 (1H, s, H-5), 6.44 (2H, s, NH_2 exchangeable with D_2O), 7.14 (2H, s, NH_2 exchangeable with D_2O), 7.65 (2H, d, *J* = 8.9 Hz, ArH), 7.92 (2H, d, *J* = 8.9 Hz, ArH), 9.34 (1H, s, NH exchangeable with D_2O) . δ_C (75 MHz, d_6-DMSO): 14.9, 61.4, 79.2, 117.9, 126.7, 135.8, 144.8, 159.3, 166.1, 170.1 . MS (ESI^+) *m*/*z* 310.25 $[M+H]^+$.HRMS (ESI+) *m/z*: Calc. for $C_{12}H_{15}N_5O_3S$, 310.0968. Found 310.0970.

General Procedure F

The chosen alcohol (10 eq.) was added dropwise to a suspension of sodium hydride (5 eq.) in THF (anh; 5 mL), and the mixture was stirred at rt under N_2 for 30 min. The chosen 4-amino-6-chloro-2-(arylamino)-5-formylpyrimidine (1 eq.) was added slowly, in small portions and the reaction mixture was heated at reflux 16 h, the allowed to cool to rt, quenched with water (10 mL), and acidified (2 M HCl) to pH < 2, then THF (10 mL) was added, the resulting mixture was stirred overnight, neutralised (saturated aq. sodium bicarbonate), and concentrated *in vacuo.* The residue was extracted with EtOAc (4 x 30 mL) and the combined organic layers were dried (Na_2SO_4), and concentrated *in vacuo* to give a yellow-orange oil. Chromatography (silica; 40-100% EtOAc, petrol, or 10% MeOH, DCM) and recrystallisation (methanol) or HPLC purification, gave the product as a white solid.

4-Amino-6-ethoxy-2-(4-methoxyphenylamino)-5-formylpyrimidine (22b)

General procedure F: 4-amino-6-chloro-2-(4-methoxyphenylamino)-5-formylpyrimidine (**21b**) (0.20 g, 0.72 mmol), ethanol (0.42 mL, 7.2 mmol). Chromatography (silica; 50-100% EtOAc, petrol) and recrystallisation (MeOH, DCM) gave **22b** as an off-white solid (0.093 g, 45%), m.p.: 111-113 °C. UV λ_{max} (EtOH): 324 nm; IR ν_{max}/ cm^{-1}: 3271, 3110, 2915, 2833, 1702, 1532, 1475, 1215; δ_H (300 MHz, $CDCl_3$): 1.39 (3H, t, *J* = 7.1 Hz, $C\mathbf{H}_3CH_2O$), 3.79 (3H, s, OCH_3), 4.41 (2H, q, *J* = 7.1 Hz, $CH_3C\mathbf{H}_2O$), 5.69 (1H, s, NH exchangeable with D_2O), 6.86 (2H, d, *J* = 8.9 Hz, ArH), 7.14 (1H, s, NH exchangeable with D_2O), 7.43 (2H, d, *J* = 8.9 Hz, ArH), 8.72 (1H, s, NH exchangeable with D_2O), 10.01 (1H, s, CHO) ; δ_C (75 MHz, $CDCl_3$): 14.3, 55.5, 62.6, 93.8, 114.2, 122.8, 131.6, 156.4, 160.8, 164.6, 171.9, 186.6 ; MS (ESI+) *m*/*z* 289.28 $[M+H]^+$. HRMS (ESI+) *m/z*: Calc. for $C_{14}H_{16}N_4O_3$: 289.1295 $[M+H]^+$. Found 289.1299 $[M+H]^+$. Anal. Calc. for $C_{14}H_{16}N_4O_3.0.1CH_2Cl_2$: requires C, 57.06; H, 5.50; N, 18.88%; found C, 57.04; H, 5.50, N, 18.41%.

4-Amino-6-isopropoxy-2-(4-methoxyphenylamino)-5-formylpyrimidine (22c)

General procedure F: 4-amino-6-chloro-2-(4-methoxyphenylamino)-5-formylpyrimidine (**21b**) (0.20 g, 0.72 mmol), isopropanol (0.55 mL, 7.2 mmol). Chromatography (silica; 50-100% EtOAc, petrol) and recrystallisation (ethanol) gave **22c** as an off-white solid (0.082 g, 38%) m.p. 137-140 °C. UV λ_{max} (EtOH): 323 nm; IR ν_{max} 3322, 2976, 2926, 2859, 1663, 1521, 1479, 1238 cm^{-1}; δ_H (300 MHz, d_4-MeOD): 1.32 (6H, d, J = 6.2 Hz, $(C\mathbf{H_3})_2CHO$), 3.72 (3H, s, OCH_3), 5.39 (1H, m, $(CH_3)_2C\mathbf{H}O$), 6.82 (2H, d, J = 9.1 Hz, ArH), 7.49 (2H, br, NH_2), 9.81 (1H, s, CHO) ; δ_C (75 MHz, $CDCl_3$): 21.7, 55.3, 69.7, 93.8, 114.0, 122.7, 131.7, 156.3, 160.8, 164.6, 171.5, 186.5 ; MS (ESI+) *m/z* 303.35 $[M+H]^+$. HRMS (ESI+) *m/z*: Calc. for $C_{15}H_{18}N_4O_3$: 303.1452 $[M+H]^+$. Found 303.1448 $[M+H]^+$. $C_{15}H_{18}N_4O_3.0.1C_2H_5OH$: requires C, 59.48; H, 6.11; N, 18.25%; found C, 59.75; H, 6.21, N, 18.23%.

4-Amino-6-*sec*-butoxy-2-(4-methoxyphenylamino)-5-formylpyrimidine (22d)

General procedure F: 4-amino-6-chloro-2-(4-methoxyphenylamino)-5-formylpyrimidine (**21b**) (0.20 g, 0.72 mmol), *sec*-butanol (0.7 mL, 7.2 mmol). Chromatography (silica, 50-100% EtOAc, Petrol) and recrystallisation (methanol) gave **22d** as an off-white solid (0.077 g, 34%) m.p. 125 -128 °C. UV λ_{max} (EtOH): 324 nm. IR ν_{max}/ cm^{-1}: 3313, 3155, 2963, 2878, 1647, 1560, 1494, 1198. δ_H (300 MHz, d_4-MeOD): 0.91 (3H, t, J = 7.4 Hz, $CH_3CHORCH_2C\mathbf{H_3}$), 1.28 (3H, d, J = 6.2 Hz, $C\mathbf{H_3}CHORCH_2CH_3$), 1.57-1.76 (2H, m, $CH_3CHORC\mathbf{H_2}CH_3$), 3.72 (3H, s, OCH_3), 5.23 (1H, m, $CH_3C\mathbf{H}ORCH_2CH_3$), 6.81 (2H, d, J = 9.2 Hz, ArH), 7.46 (2H, br, NH_2), 9.82 (1H, s, CHO) . δ_C (75 MHz, d_4-MeOD): 9.9, 19.7, 30.0, 56.1, 75.6, 94.6, 115.1, 124.3, 133.6, 157.8, 162.5, 166.3, 173.3, 187.4 . MS (ESI+) *m/z* 317.24 $[M+H]^+$. HRMS (ESI+) *m/z*: Calc. for $C_{16}H_{20}N_4O_3$: 317.1608 $[M+H]^+$. Found 317.1608 $[M+H]^+$. $C_{16}H_{20}N_4O_3$.0.4 M MeOH requires C, 60.44; H, 6.61; N, 17.09%; found C, 60.65; H, 6.15, N, 16.77%.

4-(4-amino-6-(cyclohexylmethoxy)-5-formylpyrimidin-2-ylamino)benzoic acid. (22e)

General procedure F: neopentyl 4-(4-amino-6-chloro-5-formylpyrimidin-2-ylamino)benzoate (**21c**) (0.50 g, 1.38 mmol), cyclohexylmethanol (1.7 mL, 13.8 mmol). After cooling to rt, the reaction was quenched with water (5 mL), basified to pH > 12 (2M NaOH), extracted with EtOAc (3 x 30 mL), then acidified to pH < 2 (4M HCl), and extracted with EtOAc (3 x 40 mL). The combined organic fractions were washed with water (2 x 20 mL), dried (Na_2SO_4) and concentrated *in vacuo*. Recrystallisation (EtOAc) gave **22e** as an off-white solid (0.27 g, 53%). m.p.: >250 °C dec. UV λ_{max} (EtOH): 331 and 229 nm; IR ν_{max} 3475, 3297, 3107, 2922, 2851, 1679, 1586, 1514, 1407, 1342 cm^{-1}; δ_H (300 MHz, d_6-DMSO): 1.02-1.29 (6H, m, C_6H_{11}), 1.64-1.81 (5H, m, C_6H_{11}), 4.23 (2H, d, J = 6.0 Hz, OCH_2), 7.85 (2H, d, J = 9.0 Hz, ArH), 7.95 (2H, d, J = 9.0 Hz, ArH), 8.47 (1H, br, NH), 9.96 (1H, s, CHO), 10.01 (1H, br, NH), 12.63 (1H, br, COOH) ; δ_C (75 MHz, d_6-DMSO): 25.6, 26.3, 29.6, 37.2, 71.6, 93.4, 119.7, 124.9, 130.3, 144.2, 160.5, 164.4, 167.3, 171.9, 185.8 ; MS (ESI+) *m/z* 371.30 $[M+H]^+$. HRMS (ESI+) *m/z*: Calc. for $C_{19}H_{22}N_4O_4$: 371.1714 $[M+H]^+$. Found 371.1714 $[M+H]^+$.

4-(4-Amino-6-ethoxy-5-formylpyrimidin-2-ylamino)benzenesulfonamide (22f)

General procedure F: 4-(4-amino-6-chloro-5-formylpyrimidin-2-ylamino)benzenesulfonamide (**21a**) (0.40 g, 1.22 mmol), ethanol (1.4 mL, 24.5 mmol). Chromatography (silica; 2-15% MeOH, DCM) and HPLC, Phenomenex Synergi (35-100% MeCN, water), gave **22f** as a white solid (74 mg, 9%), m.p. 266 -270 °C. UV λ_{max} (EtOH): 300 nm; IR ν_{max} 3329, 3302, 3165, 2921, 2851, 1634, 1578, 1531, 1308, 1147 cm^{-1}; δ_H (300 MHz, d_6-DMSO): 1.37 (3H, t, J = 7.0 Hz, $C\mathbf{H_3}CH_2O$), 4.47 (2H, q, J = 7.0 Hz, $CH_3C\mathbf{H_2}O$), 7.25 (2H, s, NH_2 ex), 7.72 (2H, d, J = 8.8 Hz, ArH), 7.92 (1H, s, NH ex), 7.99 (2H, d, J = 8.8 Hz, ArH), 8.48 (1H, s, NH ex), 9.95 (1H, s, CHO), 10.0 (1H, s, NH ex) ; δ_C (75 MHz, d_6-DMSO): 13.9, 62.0, 92.7, 119.1, 125.9, 137.1, 142.3, 159.7, 163.6, 170.9, 185.2 pm; MS (ESI+) *m/z* 338.1 $[M+H]^+$. HRMS (ESI+) *m/z*: Calc. for $C_{13}H_{15}N_5O_4S$: 338.0918 $[M+H]^+$. Found 338.0915 $[M+H]^+$. $C_{13}H_{15}N_5O_4S$ requires C, 46.28; H, 4.48; N, 20.76%; found C, 46.09; H, 4.47, N, 20.36%.

4-(4-Amino-5-formyl-6-isopropyloxypyrimidin-2-ylamino)benzenesulfonamide (22g)

General procedure F: 4-(4-amino-6-chloro-5-formylpyrimidin-2-ylamino)benzenesulfonamide (**21a**) (0.80 g, 2.45 mmol, 1 eq.), isopropanol (3.1 mL, 24.5 mmol). Chromatography (silica; 2-15% MeOH, DCM), and HPLC Phenomenex Synergi (35-100% MeCN, water), gave **22g** as a white solid (103 mg, 12%). m.p. 236 -240 °C. UV λ_{max}

(EtOH): 326 nm; IR ν_{max} 3290, 3209, 3078, 2984, 1641, 1566, 1312, 1149 cm^{-1}; δ_H (300 MHz, d_6-DMSO): 1.42 (6H, d, *J* = 6.2 Hz, $(CH_3)_2CHO$), 5.52 (1H, m, $(CH_3)_2CHO$), 7.84 (2H, d, *J* = 8.8 Hz, ArH), 7.97 (2H, d, *J* = 8.8 Hz, ArH), 9.97 (1H, s, CHO) ; δ_C (75 MHz, d_6-DMSO): 21.4, 69.2, 92.8, 119.1, 125.9, 137.3, 142.3, 159.7, 163.7, 170.6, 185.3 ; MS (ESI+) *m/z* 352 $[M+H]^+$. HRMS (ESI+) *m/z*: Calc. for $C_{14}H_{17}N_5O_4S$: 352.1074 $[M+H]^+$. Found 352.1070 $[M+H]^+$.

(*R*)-4-(4-Amino-6-*sec*-butoxy-5-formylpyrimidin-2-ylamino)benzenesulfonamide (22i)

General procedure F: 4-(4-amino-6-chloro-5-formylpyrimidin-2-ylamino)benzenesulfonamide (**21a**) (0.40 g, 1.22 mmol), (*R*)-*sec*-butanol (1.1 mL, 12.2 mmol). Chromatography (silica; 2-15% MeOH, DCM) and HPLC, Phenomenex Synergi (35-100% MeCN, water) gave **22i** as a white solid (36 mg, 8%), m.p.: 264-266 °C; UV λ_{max} (EtOH): 322 nm; IR ν_{max} 3288, 3218, 3154, 2974, 2877, 1646, 1566, 1483, 1409, 1386, 1155 cm^{-1}; δ_H (300 MHz, d_4-MeOD): 1.01 (3H, t, *J* = 7.4 Hz, $CH_3CHORCH_2CH_3$), 1.38 (3H, d, *J* = 6.2 Hz, $CH_3CHORCH_2CH_3$), 1.65-1.89 (2H, m, $CH_3CHORCH_2CH_3$), 5.28-5.40 (1H, m, $CH_3CHORCH_2CH_3$), 7.81 (2H, d, *J* = 8.9 Hz, ArH), 7.95 (2H, d, *J* = 8.9 Hz, ArH), 9.97 (1H, s, CHO) ; δ_C (75 MHz, d_4-MeOD): 9.8, 19.7, 29.9, 76.0, 89.7, 120.9, 127.9, 136.7, 144.3, 155.6, 162.1, 172.3, 188.1 pm; MS (ESI+) *m/z* 366.23 $[M+H]^+$. HRMS (ESI+) *m/z*: Calc. for $C_{15}H_{19}N_5O_4S$: 366.1231 $[M+H]^+$. Found 366.1234 $[M+H]^+$.

4-(4-Amino-6-cyclohexylmethoxy-5-formylpyrimidin-2-ylamino)benzenesulfonamide (22k)

General procedure F: 4-(4-amino-6-chloro-5-formylpyrimidin-2-ylamino)benzenesulfonamide (**21a**) (0.40 g, 1.22 mmol), cyclohexylmethanol (3.1 mL, 24.5 mmol). Chromatography (silica; 2-15% MeOH, DCM) and HPLC, Phenomenex Synergi (35-100% MeCN, water), gave **22k** as a white solid (0.208 g, 21%) m.p. 211-213 °C. UV λ_{max} (EtOH): 297 nm; IR ν_{max} 3302, 3147, 2922, 2849, 1645, 1576, 1524, 1307, 1141 cm^{-1}; δ_H (300 MHz, d_4-MeOD): 1.02-1.33 (6H, m, C_6H_{11}), 1.67-1.84 (5H, m, C_6H_{11}), 4.23 (2H, d, *J* = 6.1 Hz, OCH_2), 7.77 (2H, d, *J* = 8.9 Hz, ArH), 7.91 (2H, d, *J* = 8.9 Hz, ArH), 9.93 (1H, s, CHO) ; δ_C (75 MHz, d_4-MeOD): 26.8, 27.6, 30.9, 38.8, 73.2, 94.9, 120.8, 128.0, 138.3, 144.3, 157.3, 162.2, 173.7, 187.7 ; MS (ESI+) *m/z* 406.24 $[M+H]^+$; HRMS (ESI+) *m/z*: Calc. for $C_{18}H_{23}N_5O_4S$: 406.1544 $[M+H]^+$, Found 406.1546 $[M+H]^+$. $C_{18}H_{23}N_5O_4S$ requires C, 53.32; H, 5.72; N, 17.27%; found C, 53.16; H, 5.45, N, 17.01%.

4-(4-Amino-6-(1-cyclohexylethoxy)-5-formylpyrimidin-2-y-lamino)benzenesulfonamide (22l)

General procedure F: 4-(4-amino-6-chloro-5-formylpyrimidin-2-ylamino)benzenesulfonamide (**21a**) (0.40 g, 1.22 mmol), 1-cyclohexylethanol (1.2 mL, 12.2 mmol). Chromatography (silica; 2-15% MeOH, DCM) and HPLC, Waters Xterra (50-100% MeCN, water) gave **22l** as a white solid (77 mg, 15%), m.p. 185-188 °C. UV λ_{max} (EtOH): 327 nm; IR ν_{max} 3347, 3112, 2917, 2847, 1638, 1601, 1519, 1316, 1155 cm^{-1}; δ_H (300 MHz, d_4-MeOD): 0.99-1.23 (6H, m, C_6H_{11}), 1.27 (3H, d, *J* = 6.3 Hz CH_3CHOR), 1.57-1.84 (5H, m, C_6H_{11}), 5.17 (1H, m, CH_3CHOR), 7.72 (2H, d, *J* = 9.0 Hz, ArH), 7.84 (2H, d, *J* = 9.0 Hz, ArH), 9.87 (1H, s, CHO); δ_C (75 MHz, d_4-MeOD): 17.2, 27.2, 27.6, 29.9, 44.4, 78.4, 95.1, 120.9, 128.0, 138.8, 145.1, 157.3, 166.2, 173.5, 188.0 ; MS (ESI+) *m/z* 420.45 $[M+H]^+$; HRMS (ESI+) *m/z*: Calc. for $C_{19}H_{25}N_5O_4S$: 420.1700 $[M+H]^+$. Found 420.1700 $[M+H]^+$. $C_{19}H_{25}N_5O_4S$ requires C, 54.40; H, 6.01; N,16.69 %; found C, 54.53; H, 5.80; N, 16.38%.

General Procedure G

The appropriate O^4-substituted pyrimidine (1 eq.) was dissolved in 30% aq. acetic acid (1 ml – 4 ml) at rt. The temperature was increased to 80 °C and sodium nitrite (1.3 eq.) in water (1 ml – 3 ml) was added dropwise to give a purple precipitate. The reaction was maintained at 80 °C for 40 min, allowed to cool to rt and the precipitate was collected by filtration. The crude product was purified by recrystallisation from the appropriate solvent.

6-Ethoxy-5-nitrosopyrimidine-2,4-diamine (4b)

General Procedure G: ethoxypyrimidine-2,4-diamine (**3b**) (0.20 g, 1.3 mmol), 30% aq. acetic acid (1.92 mL) sodium nitrite (0.13 g, 1.8 mmol.) in water (0.29 mL). Purple crystals were collected by filtration, washed with distilled water and EtOAc and dried *in vacuo* over P_2O_5 to give **4b** as a violet powder (0.18 g, 75%) m.p. **>** 230 °C (dec). UV λ_{max}(EtOH): 334 nm; IR υ_{max} 3380, 3098, 1611, 1555, 1501 cm^{-1}; δ_H (300 MHz, d_6-DMSO): 1.40 (3H, t, *J* = 7.1 Hz,

C**H**$_3$), 4.54 (2H, q, *J* = 7.1 Hz, C**H**$_2$), 7.80 (2H, br d, *J* = 11.3 Hz, NH$_2$ ex), 8.00 (1H, s, NH ex), 10.09 (1H, s, NH ex) ; ^{13}C-NMR (75 MHz, d_6-DMSO) δ = 14.6, 63.1, 140.0, 151.5, 163.8, 170.8; MS (ESI+) *m*/*z* 184 $[M+H]^+$.

6-Cyclopropylmethoxy-5-nitrosopyrimidine-2,4-diamine (4c)

General Procedure G: 6-cyclopropylmethoxypyrimidine-2,4-diamine (**3c**) (0.44 g, 2.4 mmol), 30% aq. acetic acid (3.6 mL) sodium nitrite (0.23 g, 3.4 mmol.) in water (0.6 mL). Purple crystals were collected by filtration, washed with distilled water and EtOAc and dried *in vacuo* over P_2O_5 to give **4c** as a violet powder (0.10 g, 19%) m.p. **>** 230 °C (dec). UV λ_{max}(EtOH): 333 nm; IR υ_{max} 3287, 2963, 1613, 1572, 1510 cm^{-1}; δ_H (300 MHz, d_6-DMSO): 0.37-0.41 (2H, m, C**H**$_2$), 0.57-0.63 (2H, m, C**H**$_2$), 1.32-1.34 (1H, m, C**H**), 4.33 (2H, d, *J* = 7.2 Hz, C**H**$_2$O), 7.79 (2H, br d, *J* = 12.1 Hz, NH$_2$ ex), 8.00 (1H, s, NH ex), 10.09 (1H, s, NH ex) ; MS (ESI+) *m*/*z* 232.8 $[M+Na]^+$.

6-*sec*-Butoxy-5-nitrosopyrimidine-2,4-diamine (4d)

General Procedure G: 6-*sec*-butoxypyrimidine-2,4-diamine (**3d**) (0.2 g, 1.1 mmol, 1 eq.), 30% aqueous acetic acid (1.5 mL), sodium nitrite (0.114 g, 1.65 mmol, 1.5 eq.) in water (0.3 mL). Recrystallisation (MeOH, DCM) gave **4d** as a violet powder (0.177 g, 76.4%) m.p. 243-246 °C. UV λ_{max}(EtOH): 235 and 343 nm; IR υ_{max} 3166, 2970, 2927, 2336, 1560, 1521, 1457, 1369, 1261 cm^{-1}; δ_H (300 MHz, d_6-DMSO): 0.94 (3H, t, *J* = 7.4 Hz, CH$_3$CHORCH$_2$C**H**$_3$), 1.36 (3H, d, *J* = 6.2 Hz, C**H**$_3$CHORCH$_2$CH$_3$), 1.74 (2H, m, CH$_3$CHORC**H**$_2$CH$_3$), 5.40 (1H, m, CH$_3$C**H**ORCH$_2$CH$_3$), 7.76 (2H, s, NH$_2$ ex), 7.99 (1H, s, NH ex), 10.11 (1H, s, NH ex) ; δ_C (75 MHz, d_6-DMSO): 9.7, 19.6, 28.8, 74.5, 140.3, 151.4, 164.1, 170.9 ; MS (ESI+) *m*/*z* 212.2 $[M+H]^+$; HRMS (ESI+) *m*/*z*: Calc. for $C_8H_{13}N_5O_2$: 212.1142 $[M+H]^+$. Found 212.1142 $[M+H]^+$. $C_8H_{13}N_5O_2$.0.08 $CHCl_3$: requires C, 44.47; H, 6.08; N, 32.08%. Found C, 44.02; H, 5.98; N, 32.38%.

2,6-Diamino-4-heptoxy-5-nitrosopyrimidine, (4e).

General Procedure G: 2,6-diamino-4-heptoxypyrimidine (**3e**)(0.19 g, 0.87 mmol), 30% acetic acid (1.6 ml), and sodium nitrite (0.08 g, 1.2 mmol) in water (1 ml). Recrystallisation (methanol), gave **4e** as purple crystals (0.19 g, 86%), m.p. 208–209 °C. UV (EtOH): λ_{max1} = 356 nm, ε = 9.68 dm^3 g^{-1} cm^{-1}; IR ν_{max} 3270, 3057 N-H; 1362 cm^{-1}; λ_{max2} = 560 nm, ε = 0.26 dm^3 g^{-1} cm^{-1}; δ_H (300 MHz, d^6-DMSO) δ 0.89 (3H, t, J = 6.5 Hz, CH$_3$), 1.30–1.39 (8H, m, 4 x CH$_2$), 1.83 (2H, m, OCH$_2$C<u>H</u>$_2$), 4.51 (2H, t, *J* = 6.6 Hz, OCH$_2$), 7.82 (1H, br s, D$_2$O exch, N^{2}H<u>H</u>), 7.86 (1H, br s, D$_2$O exch, N^2<u>H</u>H), 8.05 (1H, br s, D$_2$O exch, N^{6}H<u>H</u>), 10.12 (1H, br s, D$_2$O exch, N^6<u>H</u>H); MS (ESI)$^+$ m/z = 254 $[M+H]^+$. $C_{11}H_{19}N_5O_2$ requires C, 52.16; H, 7.56; N, 27.65; found: C, 52.2; H, 7.69; N, 28.0%.

4-(4-Amino-6-ethoxy-5-nitrosopyrimidin-2-ylamino)benzenesulfonamide (20d)

General Procedure G: 4-(4-amino-6-ethoxypyrimidin-2-ylamino)benzensulfonamide (**18c**) (0.12 g, 0.388 mmol, 1 eq.), 30% aqueous acetic acid (5 mL), sodium nitrite (32 mg, 0.465 mmol, 1.2 eq.), water (0.35 mL). HPLC, Waters XTerra (Water, Methanol) provided **20d** as a green solid (15 mg, 12%) m.p. > 235 °C (dec). UV λ_{max} (EtOH): 362 nm. IR ν_{max}/ cm^{-1}: 3323, 3047, 2924, 1620, 1540, 1384, 1342, 1149. δ_H (300 MHz, d_3-MeCN): 1.54 (3H, t, *J* = 7.1 Hz, C**H**$_3$CH$_2$O), 4.72 (2H, q, *J* = 7.1 Hz, CH$_3$C**H**$_2$O), 5.68 (2H, br, NH$_2$ exchangeable with D$_2$O), 6.69 (1H, br, NH exchangeable with D$_2$O), 7.85 (2H, d, *J* = 8.8 Hz, ArH), 7.99 (2H, d, *J* = 8.8 Hz, ArH), 8.74 (1H, br, NH ex), 10.2 (1H, br, NH exchangeable with D$_2$O) ; δ_C (75 MHz, d_6-DMSO): 14.8, 63.4, 120.8, 126.6, 138.8, 140.5, 142.2, 149.9, 160.0, 171.1 . MS (ESI+) *m/z* 339.14 $[M+H]^+$. HRMS (ESI+) m/z: Calc. for $C_{18}H_{14}N_5O_4S$ $[M+H]^+$ 339.0870 ; found 339.0869.

4-(4-Amino-6-sec-butoxy-5-nitrosopyrimidin-2-ylamino)benzenesulfonamide. (20e)

General Procedure G: 4-(4-amino-6-*sec*-butoxypyrimidin-2-ylamino)benzenesulfonamide (**18b**) (0.275 g, 0.815 mmol, 1 eq.), 30% aq. acetic acid (8.0 mL), sodium nitrite (73 mg, 1.06 mmol, 1.3 eq.), water (0.6 mL). HPLC, Waters XTerra (Water, Methanol) gave **20e** as a green solid (64 mg, 21%) m.p.: 192 -195 °C. UV λ_{max} (EtOH): 362 and 257 nm. IR ν_{max}/ cm^{-1}: 3261, 3077, 2924, 1521, 1465, 1397, 1152. δ_H (300 MHz, d_3-MeCN): 0.85 (3H, t, *J* = 7.4, CH$_3$CHORCH$_2$C**H**$_3$), 1.29 (3H, d, *J* = 6.2, C**H**$_3$CHORCH$_2$CH$_3$), 1.56-1.78 (2H, m, CH$_3$CHORC**H**$_2$CH$_3$), 5.29 (1H, m, CH$_3$C**H**ORCH$_2$CH$_3$), 5.72 (2H, br, NH$_2$ ex), 6.84 (1H, br, NH ex), 7.65 (2H, d, *J* = 8.7 Hz, ArH), 7.78 (2H, br, ArH), 8.81 (1H, br, NH ex), 10.22 (1 H, br, NH ex) . δ_C (75 MHz, d_3-MeCN): 8.9, 18.6, 28.6, 76.1, 120.4, 126.8, 137.8, 140.7,

142.2, 149.9, 160.3, 171.2 . MS (ESI+) *m/z*: 367.24 $[M+H]^+$. HRMS (ESI+) *m/z* Calc. for $C_{18}H_{23}N_5O_3$ $[M+H]^+$ 367.1188, Found 367.1186.

General Procedure H

To a solution of formylpyrimidine (**22**) (0.17 mmol) in EtOH (anh; 3 mL) was added hydroxylamine hydrochloride (0.014 g, 0.202 mmol), followed by pyridine (anh., 0.016 mL, 0.202 mmol) and the mixture was stirred 16h at rt under N_2, then concentrated *in vacuo*. The residues were diluted with water (30 mL) and extracted with EtOAc (3 x 40 mL). The combined organic layers were dried(Na_2SO_4) and concentrated *in vacuo*.

4-(4-Amino-6-cyclohexylmethoxy-5-hydroxyiminomethyl)pyrimidin-2-ylamino)benzenesulfonamide (23b)

General Procedure H: 4-(4-amino-6-cyclohexylmethoxy-5-formylpyrimidin-2-ylamino)benzenesulfonamide (**22k**) (0.05 g, 0.12 mmol). Recrystallisation (methanol) gave **23b** as a white solid (0.044 g, 85%) m.p. 134-136 °C. UV λ_{max} (EtOH): 318, 261 and 219 nm; IR ν_{max} 3410, 3315, 3127, 2921, 2851, 1570, 1514, 1445, 1309, 1146, 947 cm^{-1}; δ_H (300 MHz, d_4-MeOD): 1.03-1.37 (6H, m, C_6H_{11}), 1.69-1.83 (5H, m, C_6H_{11}), 4.13 (2H, d, *J* = 6.0 Hz, OCH_2), 7.76 (2H, d, *J* = 9.0 Hz, ArH), 7.87 (2H, d, *J* = 9.0 Hz, ArH), 8.34 (1H, s, C**H**NOH) .

δ_C (75 MHz, d_4-MeOD): 26.8, 27.6, 30.9, 38.8, 73.0, 87.2, 119.5, 128.0, 136.8, 145.6, 146.1, 159.4, 163.6, 169.4 ; MS (ESI+) *m/z* 421.39 $[M+H]^+$; HRMS (ESI+) *m/z*: Calc. for $C_{18}H_{24}N_6O_4S$: 421.1653 $[M+H]^+$. Found 421.1645 $[M+H]^+$.

4-(4-Amino-6-*sec*-butoxy-5-(hydroxyiminomethyl)pyrimidin-2-ylamino)benzenesulfonamide (23c)

General Procedure H: 4-(4-amino-6-*sec*-buthoxy-5-formylpyrimidin-2-ylamino)benzenesulfonamide (**22h**) (20 mg, 54.8 µmol, 1 eq.). HPLC, Waters XTerra (35-100% MeCN, water) gave **23c** as a light yellow solid (15 mg, 83%), m.p.: 200-204 °C. UV λ_{max} (EtOH): 316 nm; IR ν_{max}3466, 3317, 3027, 2972, 1616, 1562, 1517, 1448, 1309, 1136, 951 cm^{-1}; δ_H (300 MHz, d_4-MeOD): 0.99 (3H, t, *J* = 7.4 Hz, $CH_3CHORCH_2C\mathbf{H_3}$), 1.35 (3H, d, *J* = 6.2 Hz, $C\mathbf{H}_3CHORCH_2CH_3$), 1.64-1.82 (2H, m, $CH_3CHORC\mathbf{H_2}CH_3$), 5.21-5.29 (1H, m, $CH_3C\mathbf{H}ORCH_2CH_3$), 7.78 (2H, d, *J* = 8.9 Hz, ArH), 7.91 (2H, d, *J* = 8.9 Hz, ArH), 8.35 (1H, s, C**H**NOH) ; δ_C (75 MHz, d_6-MeOD): 9.9, 19.8, 30.1, 75.3, 87.4, 119.5, 128.1, 136.8, 145.7, 146.2, 159.6, 163.8, 169.1 ; MS (ESI+) *m/z* 381.39 $[M+H]^+$; HRMS (ESI+) m/z: Calc. for $C_{15}H_{20}N_6O_4S$: 381.1340 $[M+H]^+$; found 381.1339 $[M+H]^+$.