**Supporting Information**

# Rapid Evolution of 6-Phenylpurine Inhibitors of Protein Kinase B through Structure-based Design

Alastair Donald[‡], Tatiana McHardy[‡], Martin G. Rowlands[‡], Lisa-Jane K. Hunter[‡], Thomas G. Davies[§] , Valerio Berdini[§], Robert G. Boyle[§] , Wynne G. Aherne[‡], Michelle D. Garrett[‡] and Ian Collins[*,‡]

**Contents of Supporting Information:**



[*] Corresponding author: Ian.Collins@icr.ac.uk; T +44 2087224317; F +44 02087224126

## 1. Experimental procedures for the preparation of compounds **13**, **14**, **16**, **18**, **20**.

### General Experimental

Starting materials and solvents were purchased from commercial suppliers and were used without further purification. Flash silica chromatography was performed using Merck silica gel 60 (0.025-0.04 mm). Ion exchange chromatography was performed using Isolute Flash SCX-II (acidic) or Flash NH2 (basic) resin cartridges. $^1$H NMR spectra were recorded on a Bruker AC250 or Bruker AMX500 instruments and chemical shifts (δ) are reported relative to TMS and referenced to the solvent in which they were measured. Combined HPLC-MS analyses were recorded using a Waters Alliance 2795 Separations Module and Waters/Micromass LCT mass detector with electrospray ionisation. Analytical HPLC was performed using a Supelco DISCOVERY $C_{18}$ 50 mm x 4.6 mm i.d., 5 μm column, with gradient elution of 10 to 90% MeOH / 0.1% aqueous formic acid at a flow rate of 1 mL min$^{-1}$ and a run time of 10 min. Compounds were detected at 254 nm using a Waters 2487 Dual λ Absorbance Detector. High resolution mass spectra were measured on a Waters/Micromass LCT ESI-ToF instrument, and are within +/- 5 ppm.

**(4-(9*H*-Purin-6-yl)phenyl)(phenyl)methanamine (13):** A solution of 6-chloro-9-(tetrahydro-2*H*-pyran-2-yl)-9*H*-purine (**3**)[17] (0.591 g, 2.50 mmol), 4-formylphenyl boronic acid (0.504 g, 3.42 mmol) and $(PPh_3)_4Pd$ (0.14 g, 5 mol%) in DME (24 mL) was degassed and flushed with argon. 2M aqueous $K_2CO_3$ (3.2 mL) was added and the mixture was stirred at 85 $^o$C for 18 h. The brown solution was cooled and filtered through celite, washing with EtOAc. The filtrate was concentrated and purified by flash silica column chromatography, eluting with 3:2 EtOAc:hexane, give 4-(9-(tetrahydro-2*H*-pyran-2-yl)-9*H*-purin-6-yl)benzaldehyde (**4**) (0.380 g, 1.23 mmol, 49%). $^1$H NMR ($CDCl_3$, 250 MHz) δ 1.65-2.16 (6H, m), 3.71-3.81 (1H, m), 4.04-4.18 (1H, m), 5.70-5.84 (1H, m), 8.00 (2H, dd, *J* = 2, 7 Hz), 8.32 (1H, s), 8.92 (2H, dd, *J* = 2, 7 Hz), 9.01 (1H, s), 10.06 (1H, s); *m/z* (ESI) 225 [(M+H-THP)$^+$].

Pyridinium *p*-toluenesulfonate (6 mg, 0.025 mmol), anhydrous $MgSO_4$ (0.140 g, 1.16 mmol) and **4** (0.200 g, 0.670 mmol) were added to a solution of racemic *tert*-

butanesulfinamide (0.105 g, 0.870 mmol) in dry $CH_2Cl_2$ (3.4 ml). The mixture was stirred at room temperature under nitrogen for 48 h. The mixture was filtered through celite, washing with $CH_2Cl_2$, and the filtrate was concentrated. The crude product was purified by flash silica column chromatography, eluting with 6.5:3.5 EtOAc:hexane, to give (E)-2-methyl-*N*-(4-(9-(tetrahydro-2*H*-pyran-2-yl)-9*H*-purin-6-yl)benzylidene)-propane-2-sulfinamide (**12**) as a white solid (0.124 g, 0.300 mmol, 45%). $^1$H NMR ($CDCl_3$, 250 MHz) δ 1.33 (9H, s), 1.65-2.24 (6H, m), 3.75-3.86 (1H, m), 4.13-4.28 (1H, m), 5.91-5.95 (1H, m), 8.10 (2H, d, *J* = 8 Hz), 8.48 (1H, s), 8.71 (1H, s), 9.00 (2H, d, *J* = 8 Hz), 9.17 (1H, s); *m/z* (ESI) 411 [M+H$^+$].

PhMgBr (0.06 ml, 3M solution in $Et_2O$, 0.18 mmol) was added dropwise at -60 $^o$C to a stirred solution of **12** (0.037 g, 0.090 mmol) in dry $CH_2Cl_2$ (1 mL). After 1 h the temperature was increased slowly to 0 °C. After a further 2 h, the reaction was quenched with saturated aqueous $NH_4Cl$ (1.0 mL) and extracted with EtOAc (5 mL). The organic layer was dried ($MgSO_4$) and concentrated. The crude product was purified by flash silica column chromatography, eluting with 4:1 EtOAc:hexane, to give 2-methyl-*N*-(phenyl(4-(9-(tetrahydro-2*H*-pyran-2-yl)-9*H*-purin-6-yl)phenyl)-methyl)propane-2-sulfinamide (0.017 g, 0.034 mmol, 38%). $^1$H NMR ($CD_3OD$, 250 MHz) δ 1.31 (9H, s), 1.71-2.29 (6H, m), 3.80-3.88 (1H, m), 4.11-4.20 (1H, m), 5.74 (1H, s), 5.90-5.94 (1H, m), 7.29-7.41 (3H, m), 7.51 (2H, d, *J* = 7 Hz), 7.63 (2H, d, *J* = 8 Hz), 8.66-8.70 (3H, m), 8.97 (1H, s); *m/z* (ESI) 406 [M+H$^+$-THP].

A solution of 2-methyl-*N*-(phenyl(4-(9-(tetrahydro-2*H*-pyran-2-yl)-9*H*-purin-6-yl)phenyl)methyl)propane-2-sulfinamide (0.016 g, 0.033 mmol) and 1M HCl (1 mL) in EtOH (1.3 mL) was stirred overnight at room temperature. The solution was concentrated and the crude product was purified by filtration through an Isolute-$NH_2$ basic resin cartridge (2 g), eluting with MeOH, to give **13** (0.0053 g, 0.018 mmol, 53%). $^1$H NMR ($CD_3OD$, 250 MHz) 5.30 (1H, s), 7.23-7.46 (5H, m), 7.61 (2H, d, *J* = 8 Hz), 8.45-8.52 (3H, m), 8.88 (1H, s); $^{13}$C NMR ($CD_3OD$, 125 MHz) δ 60.6, 128.15, 128.3, 128.4, 129.6, 130.8, 135.1, 145.8, 148.0, 149.2, 152.8, 154.5; *m/z* (ESI) 285 [(M-$NH_3$)$^+$]. HRMS [M+H$^+$] calcd. for $C_{18}H_{16}N_5$ 302.1406; found 302.1408. HPLC $R_t$ 4.19 min; purity (AUC) >99%.

**1-(4-(9*H*-purin-6-yl)phenyl)-2-phenylethanamine (14):** Benzylmagnesium chloride (0.36 mL, 2M solution in THF, 0.72 mmol) was added dropwise at room temperature

to a stirred solution of **12** (0.097 g, 0.23 mmol) in dry THF (8 mL). The solution was refluxed under $N_2$ for 4 h. The reaction was cooled, diluted with saturated aqueous $NH_4Cl$ (10 mL) and extracted with EtOAc (2 x 10 mL). The organic layer was dried ($MgSO_4$) and concentrated. The crude product was purified by preparative TLC, eluting with 4:1 EtOAc/hexane, to give 2-methyl-*N*-(2-phenyl-1-(4-(9-(tetrahydro-2*H*-pyran-2-yl)-9*H*-purin-6-yl)phenyl)ethyl)propane-2-sulfinamide (0.038 g, 0.076 mmol, 33%). $^1$H NMR ($CD_3OD$, 250 MHz) δ 1.22 (9H, s), 1.72-2.34 (6H, m), 3.13-3.22 (2H, m), 3.82-3.89 (1H, m), 4.16-4.20 (1H, m), 4.67-4.73 (1H, m), 5.89-5.95 (1H, m), 7.12-7.49 (7H, m), 8.62-8.68 (3H, m), 8.97 (1H, s); *m/z* (ESI) 420 [(M+H-THP)$^+$].

A solution of 2-methyl-*N*-(2-phenyl-1-(4-(9-(tetrahydro-2*H*-pyran-2-yl)-9*H*-purin-6-yl)phenyl)ethyl)propane-2-sulfinamide (0.038 g, 0.076 mmol) and 4M HCl in 1,4-dioxane (0.2 mL) in MeOH (1.5 mL) was stirred at room temperature for 16 h. The solution was concentrated and the crude product was purified by filtration through an Isolute-$NH_2$ basic resin cartridge (2 g), eluting with MeOH, to give **14** (0.016 mg, 0.051 mmol, 67%). $^1$H NMR ($CD_3OD$, 250 MHz) δ 2.97 (2H, d, *J* = 7 Hz), 4.17 (1H, t, *J* = 7 Hz), 7.02-7.17 (5H, m), 7.40 (2H, d, *J* = 8 Hz), 8.32-8.39 (3H, m), 8.75 (1H, s); *m/z* (ESI) 316 [(M+H)$^+$]. HRMS [M+H$^+$] calcd. for $C_{19}H_{18}N_5$ 316.1562; found 316.1562. HPLC $R_t$ 4.35 min; purity (AUC) >99 %.

**(4-(9H-Purin-6-yl)phenyl)(4-chlorophenyl)methanamine (16):** 4-Chlorophenyl magnesium bromide (40 mL, 1M solution in $Et_2O$, 40 mmol) was added dropwise to a solution of 4-bromobenzaldehyde (6.90 g, 37 mmol) in THF (20 mL) at 0 $^o$C. The solution was stirred for 50 minutes, and then saturated aqueous $NH_4Cl$ (200 mL) and EtOAc (250 mL) were added. The organic layer was washed with water (100 mL), dried ($Na_2SO_4$), concentrated and purified by flash silica column chromatography, eluting with 6:1 hexane:EtOAc, to give 1-((4-bromophenyl)-(4-chlorophenyl))-methanol (**15**) (4.47 g, 41%). $^1$H NMR ($(CD_3)_2SO$, 250 MHz) δ 5.71 (1H, s), 6.05 (1H, br s), 7.31-7.38 (6H, m), 7.51 (2H, d, *J* = 8 Hz). *m/z* (ESI) 279 [M+H$^+$-$H_2O$]. Diisopropylazodicarboxylate (2.40 mL, 12.19 mmol) was added dropwise to a solution of **15** (2.30 g, 7.73 mmol), $PPh_3$ (3.42 g, 13.03 mmol) and phthalimide (1.91 g, 12.98 mmol) in THF (60 mL). The solution was stirred for 18 h, and then poured into $Et_2O$ (250 mL). The solution was washed with saturated aqueous $NaHCO_3$ (2 x

100 mL) and brine (50 mL). The organic fraction was dried ($Na_2SO_4$), concentrated, and purified by flash silica column chromatography, eluting with 6:1 hexane:EtOAc, to give 2-((4-bromophenyl)(4-chlorophenyl)methyl)isoindoline-1,3-dione (0.70 g, 21%). $^1$H NMR ($CDCl_3$, 250 MHz) δ 6.64 (1H, s), 7.24-7.33 (6H, m), 7.50 (2H, d, *J* = 8 Hz), 7.77 (2H, dd, *J* = 6, 3 Hz), 7.88 (2H, dd, *J* = 6, 3 Hz); *m/z* (ESI) 426 [$M+H^+$].

A solution of $Pd_2(dba)_3$ (13 mg, 0.014 mmol) and tricyclohexylphosphine (20 mg, 0.07 mmol) in 1,4-dioxane (6 mL) was degassed and stirred at room temperature for 30 min. Bis(pinacolato)diboron (0.256 g, 1 mmol), 2-((4-bromophenyl)(4-chlorophenyl)methyl)isoindoline-1,3-dione (0.424 g, 1 mmol) and KOAc (0.164 g, 1.67 mmol) were added, and the solution was heated at 80 °C for 16 h. After cooling to room temperature, the solution was diluted with EtOAc (10 mL) and washed with water (50 mL) and brine (50 mL). The organic layer was dried ($Na_2SO_4$), concentrated and purified by flash silica column chromatography, eluting with 6:1 hexane:EtOAc, to give 2-((4-chlorophenyl)(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)methyl)isoindoline-1,3-dione (0.142 g, 30%). 1H NMR ($CDCl_3$, 250 MHz) δ 1.35 (12H, s), 6.70 (1H, s), 7.33-7.38 (6H, m), 7.74-7.89 (6H, m); (ESI) m/z (ESI) 497 [$M+Na^+$].

$PdCl_2(PPh_3)_2$ (22 mg, 7 mol%) and 1M aqueous $K_2CO_3$ (1 mL) were added to a solution of 6-chloro-9-(tetrahydro-2*H*-pyran-2-yl)-9*H*-purine (**3**) (0.105 g, 0.44 mmol) and 2-((4-chlorophenyl)-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-phenyl)-methyl)-isoindoline-1,3-dione (0.211 mg, 0.44 mmol) in DME (2 mL). The solution was heated at 80 °C for 18 h. The mixture was diluted with water (50 mL) and extracted with chloroform (2 x 100 mL). The organic extracts were dried ($Na_2SO_4$), concentrated, and purified by flash silica column chromatography, eluting with 1:1 then 1:3 hexane:EtOAc, to give 2-((4-chlorophenyl)(4-(9-(tetrahydro-2*H*-pyran-2-yl)-9*H*-purin-6-yl)phenyl)methyl)isoindoline-1,3-dione (0.101 g, 42%). $^1$H NMR ($CDCl_3$, 250 MHz) δ 1.60-2.30 (6H, m), 3.82 (1H, dt, *J* = 3, 11 Hz), 4.15-4.26 (1H, m), 5.85 (1H, dd, *J* = 3,10 Hz), 6.77 (1H, s), 7.30-7.41 (4H, m), 7.55 (2H, d, *J* = 8 Hz), 7.74 (2H, dd, *J* = 3, 5Hz), 7.86 (2H, dd, *J* = 3, 6 Hz), 8.33 (1H, s), 8.76 (2H, d, *J* = 8 Hz), 9.01 (1H, s).

Hydrazine hydrate (1 mL) was added to a solution of 2-((4-chlorophenyl)(4-(9-(tetrahydro-2*H*-pyran-2-yl)-9*H*-purin-6-yl)phenyl)methyl)isoindoline-1,3-dione (0.099 g, 0.18 mmol) in EtOH (6 mL). The solution was stirred for 48 h. The resulting

precipitate was removed by filtration and the filtrate was concentrated. The residue obtained was dissolved in methanol and purified by ion exchange on SCX-II acidic resin, eluting with MeOH and then with 3M $NH_3$ in MeOH. The basic fractions were combined and the crude material was redissolved in MeOH (2 mL). 4M HCl in 1,4-dioxane (2 mL) was added and the mixture was stirred for 18 h. The mixture was concentrated. The residues were redissolved in MeOH and purified by ion exchange on SCX-II acidic resin, eluting with MeOH and then with 3M $NH_3$ in MeOH. The basic fractions were concentrated to give **16** (0.044 g, 73% over 2 steps). $^1$H NMR ($(CD_3)_2SO$, 250 MHz) δ 5.22 (1H, s), 7.36 (2H, d, *J* = 8 Hz), 7.48 (2H, d, *J* = 8 Hz), 7.62 (2H, d, *J* = 8 Hz), 8.62 (1H, s), 8.73 (2H, d, *J* = 8 Hz), 8.93 (1H, s); *m/z* (ESI) 336 [$M+H^+$]. HRMS $M+H^+$ calcd. for $C_{18}H_{15}ClN_5$ 336.1016; found 336.1012. HPLC $R_t$ 4.11 min; purity (AUC) >99%.

**2-(4-(9*H*-Purin-6-yl)phenyl)-2-(4-chlorophenyl)-*N*-methylethanamine (18):** $AlCl_3$ (2.80 g, 21 mmol) was added in small portions to a solution of 1-(4-bromophenyl)-2-methylamino-ethanol (2.04 g, 8.87 mmol) in chlorobenzene (15 mL) at 0 $^o$C. After addition was complete, the mixture was warmed to room temperature and stirred for 5 h, then poured onto ice (200 mL). The mixture was diluted with 2M aqueous NaOH (100 mL) and extracted with $CH_2Cl_2$ (200 mL). The organic layer was washed with brine (50 mL), dried ($MgSO_4$) and concentrated to give 2-(4-bromophenyl)-2-(4-chlorophenyl)-*N*-methylethanamine (**17**) (2.70 g, 94%). $^1$H NMR ($CDCl_3$, 250 MHz) δ 2.47 (3H, s), 3.18 (2H, d, *J* = 8 Hz), 4.18 (1H, t, *J* = 8 Hz), 7.13 (2H, d, *J* = 8 Hz), 7.17 (2H, d, *J* = 8 Hz), 7.31 (2H, d, *J* = 8 Hz), 7.46 (2H, d, *J* = 8 Hz).

$Et_3N$ (1.5 mL, 10.7 mmol) and di-*tert*-butyl dicarbonate (1.94 g, 8.9 mmol) were added to a solution of **17** (2.70 g, 8.32 mmol) in $CH_2Cl_2$ (100 mL) and the mixture was stirred at room temperature for 18 h. The solution was diluted with $CH_2Cl_2$ (250 mL), washed with 1M aqueous citric acid (200 mL), dried ($MgSO_4$) and concentrated to give *tert*-butyl 2-(4-bromophenyl)-2-(4-chlorophenyl)ethyl(methyl)carbamate (3.77 g, 100%). $^1$H NMR ($(CD_3)_2SO$, 250 MHz) δ 1.38 (9H, s), 2.73 (3H, s), 3.90 (2H, d, *J* = 8 Hz), 4.50 (1H, br s), 7.34 (2H, d, *J* = 8 Hz), 7.35-7.44 (4H, m), 7.51 (2H, d, *J* = 8 Hz); *m/z* (ESI) 446 [$M+Na^+$].

A solution of $Pd_2(dba)_3$ (0.034 g, 0.037 mmol) and tricyclohexylphosphine (0.052 g, 0.18 mmol) in 1,4-dioxane (20 mL) was degassed and stirred at room temperature for

30 min. Bis(pinacolato)diboron (1.0 g, 3.96 mmol), *tert*-butyl 2-(4-bromophenyl)-2-(4-chlorophenyl)ethyl(methyl)carbamate (1.54 g, 3.60 mmol) and KOAc (0.58 g, 5.91 mmol) were added, and the solution was heated at 80 °C for 16 h. After cooling to room temperature, the solution was diluted with EtOAc (20 mL) and washed with water (50 mL) and brine (50 mL). The organic layer was dried ($Na_2SO_4$), concentrated and purified by flash silica column chromatography, eluting with 6:1 hexane:EtOAc, to give (2-(4-chlorophenyl)-2-[4-(4,4,5,5-tetramethyl-[1,3,2]dioxaborolan-2-yl)-phenyl]-ethyl)-methyl-carbamic acid *tert*-butyl ester (0.997 g, 2.11 mmol, 59%). 1H NMR ($(CD_3)_2CO$, 250 MHz) δ 1.34 (9H, s), 1.39 (12H, s), 2.71 (3H, br s), 3.83-4.05 (2H, m), 4.40-4.57 (1H, m), 7.32-7.45 (6H, m), 7.72 (2H, d, $J$ = 8 Hz); m/z (ESI) 494 [$M+Na^+$].

$PdCl_2(PPh_3)_2$ (0.023 g, 5mol%) and 1M aqueous $K_2CO_3$ (2 mL) were added to a solution of 6-chloro-9-(tetrahydro-2*H*-pyran-2-yl)-9*H*-purine (**3**) (0.159 g, 0.67 mmol) and (2-(4-chloro-phenyl)-2-[4-(4,4,5,5-tetramethyl-[1,3,2]dioxaborolan-2-yl)-phenyl]-ethyl)-methyl-carbamic acid *tert*-butyl ester (0.250 g, 0.53 mmol) in DME (2 mL). The solution was heated at 80 °C for 18 h. The mixture was diluted with water (50 mL) and extracted with $CH_2Cl_2$ (2 x 50 mL). The organic extracts were dried ($MgSO_4$), concentrated, and purified by flash silica column chromatography, eluting with 4:1 hexane:EtOAc, to give *tert*-butyl 2-(4-chlorophenyl)-2-(4-(9-(tetrahydro-2*H*-pyran-2-yl)-9*H*-purin-6-yl)phenyl)ethyl(methyl)carbamate (0.142 g, 0.260 mmol, 49%). $^1$H NMR ($CDCl_3$, 250 MHz) δ 1.42 (9H, s), 1.50-2.00 (3H, m), 2.00-2.20 (3H, m), 2.60-2.85 (3H, m), 3.80-4.10 (3H, m), 4.18-4.29 (1H, m), 5.88 (1H, d, $J$ = 10 Hz), 7.20-7.40 (4H, m), 7.40-7.50 (2H, m), 8.36 (1H, s), 8.75 (2H, d, $J$ = 7 Hz), 9.03 (1H, s); *m/z* (ESI) 548 [$M+H^+$].

4M HCl in 1,4-dioxane (2 mL) was added to a stirred solution of *tert*-butyl 2-(4-chlorophenyl)-2-(4-(9-(tetrahydro-2*H*-pyran-2-yl)-9*H*-purin-6-yl)phenyl)ethyl-(methyl)carbamate (0.140 g, 0.260 mmol) in MeOH (2 mL). The solution was stirred for 24 h, and then concentrated. The residue was dissolved in MeOH and purified by ion exchange on SCX-II acidic resin, eluting with MeOH and then with 3M $NH_3$ in MeOH. The basic fractions were concentrated to give **18** (0.089 g, 95%). $^1$H NMR ($d_6$-DMSO, 500 MHz) δ 2.32 (3H, s), 3.15-3.19 (2H, m), 4.27 (1H, t, $J$ = 8 Hz), 7.36 (2H, d, $J$ = 8 Hz), 7.38 (2H, d $J$ = 8 Hz), 7.51 (2H, d, $J$ = 8 Hz), 8.60 (1H, s), 8.74

(1H, d, *J* = 8 Hz), 8.91 (1H, s); *m/z* (ESI) 364 [M+$H^+$]. HRMS [M+$H^+$] calcd. for $C_{20}H_{19}ClN_5$ 364.1329; found 364.1336. HPLC $R_t$ 4.55 min; purity (AUC) 98%.

**6-(4-(4-(4-Chlorophenyl)piperidin-4-yl)phenyl)-9H-purine (20):** Di-*tert*-butyl dicarbonate (1.70 g, 7.79 mmol) was added to a solution of 4-(4-bromophenyl)-4-(4-chlorophenyl)-piperidine hydrochloride (**19**)[22] (2.80 g, 7.45 mmol) and $Et_3N$ (8.0 mL, 57.4 mmol) in $CH_2Cl_2$ (150 mL). The solution was stirred for 18 h at room temperature, then washed with 1M aqueous citric acid (100 mL). The organic layer was dried ($Na_2SO_4$) and concentrated to give *tert*-butyl 4-(4-bromophenyl)-4-(4-chlorophenyl)piperidine-1-carboxylate (3.14 g, 94%). $^1$H NMR (250 MHz, d6-acetone) δ 1.47 (9H, s), 2.37-2.47 (4H, m), 3.41-3.53 (4H, m), 7.32-7.44 (6H, m), 7.51 (2H, d, *J* = 9 Hz). *m/z* (ESI) 349 [M+$H^+$-Boc].

A solution of n-BuLi in hexanes (2M, 1.0 mL, 2.0 mmol) was added dropwise at –78 $^o$C to a stirred solution of *tert*-butyl 4-(4-bromophenyl)-4-(4-chlorophenyl)piperidine-1-carboxylate and tri(isopropyl)borate (0.47 mL, 2.0 mmol) in dry THF (6 mL) under $N_2$. After 3 h the initial orange solution had become pale yellow. The reaction was warmed to room temperature for 1 h, then quenched with 1M HCl (3 mL). The mixture was diluted with water (20 mL), 1M HCl (10 mL) and extracted with EtOAc (10 mL). The organic layer was dried ($Na_2SO_4$), filtered and concentrated to give 4-(1-(*tert*-butoxycarbonyl)-4-(4-chlorophenyl)piperidin-4-yl)phenylboronic acid as a white foam (0.15 g, 0.361 mmol, 26%). *m/z* (ESI) 452 [M+$Na^+$].

A solution of 4-(1-(*tert*-butoxycarbonyl)-4-(4-chlorophenyl)piperidin-4-yl)phenylboronic acid (0.083 g, 0.20 mmol), 6-chloro-9-(tetrahydro-2*H*-pyran-2-yl)-9*H*-purine (**3**)[16] (0.050 g, 0.21 mmol), 2M aqueous $K_2CO_3$ (0.20 mL, 0.40 mmol) and $Pd(PPh_3)_4$ (0.020 g, 7 mol%) in DME (3 mL) was flushed with $N_2$ then stirred at 85 $^o$C for 16 h. The solution was diluted with water (15 mL) and extracted with EtOAc (15 mL). The extract was dried ($Na_2SO_4$), filtered and concentrated. Preparative TLC, eluting with 1:1 EtOAc:hexanes, gave tert-butyl 4-(4-chlorophenyl)-4-(4-(9-(tetrahydro-2*H*-pyran-2-yl)-9*H*-purin-6-yl)phenyl)piperidine-1-carboxylate (0.030 g, 0.052 mmol, 26%). $^1$H NMR (250 MHz, $CDCl_3$) δ 1.38 (9H, s), 1.50-1.85 (6H, m), 1.98-2.08 (2H, m), 2.25-2.45 (4H, m), 3.30-3.50 (4H, m), 3.65-3.80 (1H, m), 4.14 (1H, d, *J* = 13 Hz), 5.78 (1H, d, *J* = 10 Hz), 7.11-7.19 (4H, m), 7.37 (2H, d, *J* = 8 Hz), 8.25 (1H, s), 8.60 (2H, d, *J* = 8 Hz), 8.93 (1H, s). *m/z* (ESI) 490 [M+$H^+$-THP].

A solution of 4-(4-chlorophenyl)-4-(4-(9-(tetrahydro-2H-pyran-2-yl)-9H-purin-6-yl)phenyl)piperidine-1-carboxylate (0.030 g, 0.052 mmol) and 1M HCl (2 mL) in EtOH (4 mL) was stood at room temperature for 24 h then warmed to 80 $^{o}$C for 5 h. The mixture was purified by ion-exchange chromatography on SCX-II acidic resin (2 g), eluting with MeOH then 1M ammonia-MeOH. The basic fractions were combined, concentrated and triturated with $Et_2O$ to give **20** as an off-white solid (0.014 g, 0.036 mmol, 69%). $^{1}$H NMR (500 MHz, $CD_3OD$) $\delta$ 2.50-2.55 (2H, m), 2.59-2.64 (2H, m) 3.04 (4H, dd, *J* = 6, 6 Hz), 3.65-3.80 (1H, m), 7.31 (2H, d, *J* = 9 Hz), 7.37 (2H, d, *J* = 9 Hz), 7.54 (2H, d, *J* = 9 Hz), 8.39 (1H, s), 8.45 (2H, d, *J* = 9 Hz), 8.82 (1H, s). *m/z* (ESI) 390 [M+H$^{+}$]. HRMS [M+H$^{+}$] calcd. for $C_{22}H_{21}ClN_5$ 364.1485; found 390.1478. HPLC $R_t$ 5.00 min; purity (AUC) >99%.

**2. Table S1: HPLC purities of compounds important for SAR**

| Compound | HPLC purity (AUC) 1 [a] | HPLC purity (AUC) 2 [b] |
|---|---|---|
| **16** | >99% | 96% |
| **18** | 98% | 93% |
| **20** | >99% | 97% |

[a] Method 1: HPLC was performed using a Supelco DISCOVERY $C_{18}$ 50 mm x 4.6 mm i.d., 5 µm column, with gradient elution of 10 to 90% MeOH / 0.1% aqueous formic acid at a flow rate of 1 mL $min^{-1}$ and a run time of 10 min. UV detection at 254 nm.

[b] Method 2: HPLC was performed using a Supelco DISCOVERY $C_{18}$ 30 mm x 4.6 mm i.d., 5 µm column, with gradient elution of 10 to 90% MeOH / 0.1% aqueous formic acid at a flow rate of 1 mL $min^{-1}$ and a run time of 6 min. UV detection at 254 nm.

### 3. PKA-PKB crystallography

X-ray structure determinations for the PKA-PKB chimera-inhibitor complexes discussed in this paper were carried out as described previously.[16] Data collection and refinement statistics are presented in Table S2. Coordinates and structure factors for the complexes have been deposited with the PDB with the following accession codes: 2UVX (PKA-PKB-**2**), 2UVY (PKA-PKB-**6**), 2UVZ (PKA-PKB-**13**) and 2UW0 (PKA-PKB-**20**).

**Table S2: X-ray data collection and refinement statistics for the PKA-PKB inhibitor complexes**

| | PKA-PKB-**2** | PKA-PKB-**6** | PKA-PKB-**13** | PKA-PKB-**20** |
|---|---|---|---|---|
| **Data collection** | | | | |
| Space group | $P2_12_12_1$ | $P2_12_12_1$ | $P2_12_12_1$ | $P2_12_12_1$ |
| Cell dimensions | | | | |
| *a*, *b*, *c* (Å) | 72.54, 74.40, 79.87 | 72.72, 74.65, 80.07 | 72.58, 74.66, 80.27 | 72.29, 74.42, 79.95 |
| α, β, γ (°) | 90, 90, 90 | 90, 90, 90 | 90, 90, 90 | 90, 90, 90 |
| Resolution (Å) | 2.00 (2.11 – 2.00) | 1.95 (1.97 – 1.95) | 1.94 (1.97 – 1.94) | 2.00 (2.07 – 2.00) |
| $R_{merge}$ [a] | 7.1 (29.4) | 4.7 (19.1) | 6.1 (30.3) | 7.5 (28.7) |
| $I / \sigma I$ | 9.6 (2.0) | 6.6 (3.4) | 6.6 (2.4) | 6.4 (2.8) |
| Completeness (%) | 97.5 (93.0) | 97.1 (87.2) | 96.8 (88.5) | 98.4 (95.1) |
| Redundancy | 2.4 (2.0) | 2.6 (2.0) | 2.4 (1.9) | 2.7 (2.4) |
| | | | | |
| **Refinement** | | | | |
| Resolution (Å) | 30.6 – 2.00 | 35.3 – 1.95 | 27.3 – 1.94 | 35.2 – 2.00 |
| No. reflections | 27518 | 29879 | 30508 | 28197 |
| $R_{work}$ [b] / $R_{free}$ [c] | 17.2/23.7 | 21.6/25.3 | 20.3/24.4 | 20.3/24.8 |
| No. atoms | | | | |
| Protein + peptide | 2941 | 2939 | 2945 | 2945 |
| Inhibitor | 9 | 18 | 23 | 28 |
| Water | 383 | 327 | 296 | 292 |
| *B*-factors (Å$^2$) | | | | |
| Protein + peptide | 23 | 25 | 29 | 28 |
| Ligand | 12 | 16 | 23 | 25 |
| Water | 32 | 30 | 32 | 34 |
| R.m.s deviations | | | | |
| Bond lengths (Å) | 0.012 | 0.006 | 0.007 | 0.007 |
| Bond angles (°) | 1.3 | 1.0 | 1.0 | 1.1 |

[a] $R_{merge} = \Sigma_h\Sigma_j |I_{h,j} - \bar{I}_h|/\Sigma_h\Sigma_j|I_{h,j}|$, where $I_{h,j}$ is the *j*th observation of reflection *h*.
[b] $R_{work} = \Sigma_h||F_{oh}| - |F_{ch}||/\Sigma_h|F_{oh}|$, where $F_{oh}$ and $F_{ch}$ are the observed and calculated structure factor amplitudes respectively for the reflection *h*.
[c] $R_{free}$ is equivalent to $R_{work}$ for a 5% subset of reflections not used in the refinement.

## 4. Experimental procedures for the GSK3β-ELISA and SRB cellular assays

### Phospho-GSK3β (Ser9) Cellular ELISA Assay

Cells were plated in 96 well microplates at 16,000 cells per well in media supplemented with 10% FBS, and grown for 24 h before treatment with compound. Compound or vehicle control was added to the cells for 1 h. Following this, cells were fixed with 3% paraformaldehyde, 0.25% glutaraldehyde, 0.25% Triton-X100 and blocked with 5% milk in TBST prior to overnight incubation with a phospho-GSK3β (serine 9) antibody (Cell Signaling Technology). The plates were then washed, secondary antibody added, and enhancement of the signal performed using DELFIA reagents (Perkin Elmer) as stated in the manufacturer's instructions. Europium counts were normalized to the protein concentration, and the $IC_{50}$ value for each inhibitor was calculated in GraphPad Prism 4.00 using non-linear regression analysis and a sigmoidal dose response (variable slope) equation.

### Sulforhodamine B (SRB) Growth Assay.

Cells were plated in 96-well microplates at 6,000 cells and 1,125 cells respectively per well in 160μl DMEM with 10% foetal bovine serum and grown for 48 hr before treating with inhibitor. Compound was added in 40 μl media per well to a final concentration ranging from 0.2–50 μM in quadruplicate wells. Control wells received media containing DMSO vehicle to the same final concentration. Following 96 hours of treatment, plates were fixed, stained and analyzed as previously described.[1] The $IC_{50}$ value for each inhibitor was calculated in GraphPad Prism 4.00 using non-linear regression analysis and a sigmoidal dose response (variable slope) equation.

[1] P. Skehan, R. Storeng, D. Scudiero, A. Monks, J. McMahon, D. Vistica, J. T. Warren, H. Bokesch, S. Kenney, M. R. Boyd. New colorimetric cytotoxicity assay for anticancer-drug screening. *J. Natl. Cancer Inst.* 1990, **82**, 1107-1112.