# Supporting Information

# The design and synthesis of potent and selective inhibitors of BRD7 and BRD9 bromodomains

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## Synthetic procedures

### List of Abbreviations

Ac – Acetate
Aq. – Aqueous
Boc – *tert*-Butoxycarbonyl
CVs – Column volumes
DAD – Diode Array Detector
Dec – Decomposition (during melting point determination)
DMF – Dimethylformamide

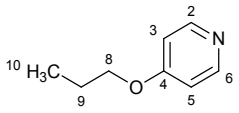
DMSO – Dimethyl sulphoxide
ESI – Electrospray Ionisation
LCMS – High Performance Liquid Chromatography
HRMS – High Resolution Mass Spectrometry
LRMS – Low Resolution Mass Spectrometry
Ph – Phenyl
MeCN – Acetonitrile
mp – Melting point
MS – Mass spectrometry
NMR – Nuclear Magnetic Resonance
$R_f$ – Retardation factor
SCX – Strong cation exchange
TBAF – Tetrabutylammonium fluoride
THF – Tetrahydrofuran
TIPS – Triisopropylsilyl
TLC – Thin Layer Chromatography
$t_r$ – Retention time

## General Experimental

All reactions involving organometallic or other moisture-sensitive reagents were carried out under a nitrogen atmosphere using standard vacuum line techniques and glassware that was oven dried and cooled under nitrogen before use. Commercial anhydrous solvents used in reactions and LCMS grade solvents were employed for work-up and chromatography. Water was purified using an Elix UV-10 system. All other reagents were used as supplied (analytical or LCMS grade) without prior purification. Parallel synthesis was carried out using a Radleys GreenHouse reactor. Parallel work-ups were carried out using a Radleys stacker and Isolute phase separation cartridges. Thin layer chromatography was performed on aluminium plates coated with 60 $F_{254}$ silica. Plates were visualised using UV light (254 nm) or 1% aq. $KMnO_4$. $R_f$ values are quoted to the nearest 0.05. Flash column chromatography was performed either on Kieselgel 60 silica on a glass column, or on a Biotage SP4 automated flash column chromatography platform. Melting points were recorded on a Gallenkamp Hot Stage apparatus. Optical rotations were recorded on a Perkin-Elmer 241 polarimeter with a water-jacketed 10 cm cell. Specific rotations are reported in $10^{-1}$ deg $cm^2$ $g^{-1}$ and concentrations in g/100 mL. IR spectra were recorded on a Bruker Tensor 27 FT-IR spectrometer; selected characteristic peaks are reported in $cm^{-1}$. NMR spectra were recorded on Bruker Avance spectrometers in the deuterated solvent stated. Spectra were recorded at room temperature unless otherwise stated. The field was locked by external referencing to the relevant deuteron resonance. Coupling constants (*J*) are quoted in Hz and are recorded to the nearest 0.5 Hz. Identical proton coupling constants are averaged in each spectrum and reported to the nearest 0.5 Hz. When peak multiplicities are reported, the following abbreviations are used: s = singlet, d = doublet, t = triplet, m = multiplet, br = broadened, dd = doublet of doublets, dt = doublet of triplets. *m/z* values are reported in Daltons. LRMS were recorded on a Waters LCT Premier, equipped with electrospray ionisation source and TOF analyser, acquiring in positive and negative ionisation modes or on and Agilent 6100 mass spectrometer operated with an electrospray ionisation source via flow injection analysis with an Agilent 1200 isocratic pump; data acquisition and processing was performed using Waters Masslynx 4.1 software or Agilent chemstation software. HRMS were run on either a Bruker MicroTOF internally calibrated with polyalanine, or a Micromass GCT instrument fitted with a

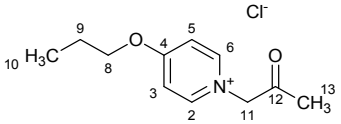
Scientific Glass Instruments BPX5 column (15 m × 0.25 mm) using amyl acetate as a lock mass. Elemental analyses were recorded by the elemental analysis service of the London Metropolitan University. Microwave experiments were carried out using a Biotage Initiator 8. Flash column chromatography was carried out using a Presearch Isco Combiflash Companion using Presearch columns or on a Biotage SP4 using Biotage SNAP columns. LCMS $t_r$ are quoted to the nearest 0.1 min. LCMS were performed on the following systems: System A: WATERS sunfire C18 column (150 mm × 4.6 mm, 5 µm) using a linear gradient of solvent A (water + 0.01% $CF_3CO_2H$) and solvent B (acetonitrile + 0.01% $CF_3CO_2H$), eluting at a flow rate of 1 mL/min and monitoring at 254 nm: 0% B over 2 min, 0% B to 100% B over 16 min and 100% B over 2 min; System B: Merk Millipore Chromolith Performance RP-18e column (100 mm × 2 mm, 1.6 µm) using a linear gradient of solvent A (water + 0.01% $CF_3CO_2H$) and solvent B (acetonitrile + 0.01% $CF_3CO_2H$), eluting at a flow rate of 1 mL/min and monitoring at 254 nm: 2% B over 2 min, 2% B to 100% B over 8 min and 100% B over 1 min.

## 4-Propoxypyridine




Sodium metal (345 mg, 15.00 mmol) was added in portions to anhydrous *n*-PrOH (8 mL) at room temperature using a water bath to dissipate evolved heat. Once all sodium had dissolved, the resulting colourless solution was added drop-wise to a suspension of 4-chloropyridine hydrochloride (750 mg, 5.00 mmol) in *n*-PrOH (2 mL). The resultant suspension was heated at reflux for 16 h. The mixture was allowed to cool, neutralised by addition of 1 M aq. HCl, and then concentrated *in vacuo*. The material thus obtained was partitioned between EtOAc (10 mL) and water (10 mL). The phases were separated then the organic phase was washed with brine (10 mL) then dried over $MgSO_4$ and concentrated *in vacuo*. The crude material was purified by flash column chromatography on a silica column (24 g). The column was eluted with a gradient of EtOAc:*c*-hexane which was increased linearly from 40:60 to 80:20 over 12 CVs. The desired fractions were combined and evaporated to yield the product as a colourless oil (637 mg, 93%); $R_f$ 0.10 (EtOAc:*c*-hexane, 40:60); $\nu_{max}$ (neat) 3031 (C-H), 2967 (C-H), 2939 (C-H), 2880 (C-H); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 1.03 (t, *J*=7.5 Hz, 3 H, C(10)$H_3$), 1.81 (sxt, *J*=7.5 Hz, C(9)$H_2$), 3.95 (t, *J*=6.5 Hz, 2 H, C(8)$H_2$), 6.74 - 6.84 (m, 2 H, C(3)*H*+C(5)*H*), 8.31 - 8.50 (m, 2 H, C(2)*H*+C(6)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 10.3 (s, 1 C, *C*(10)), 22.2 (s, 1 C, *C*(9)), 69.3 (s, 1 C, *C*(8)), 110.2 (s, 2 C, *C*(3)+*C*(5)), 150.9 (s, 2 C, *C*(2)+*C*(6)), 165.0 (s, 1 C, *C*(4)); LRMS *m/z* ($ESI^+$) 138 [$MH^+$]; HRMS ($ESI^+$) found 138.0907, calculated for $C_8H_{12}O^+$ 138.0913; HPLC (System E) $t_r$ 1.1 min (>99%).

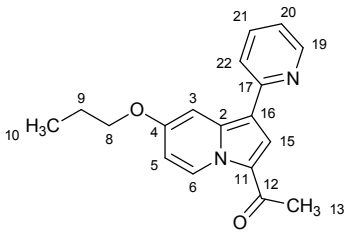
## 1-(2-Oxopropyl)-4-propoxypyridinium chloride (4)




Chloroacetone (293 µL, 3.56 mmol) was added to a stirred solution of 4-propoxypyridine (630 mg, 3.24 mmol) in THF (15 mL). The resultant mixture was left to stir for 16 h at room temperature then more chloroacetone (293 µL, 3.56 mmol) was added. The mixture was refluxed for 6 h then allowed to cool. $Et_2O$ (10 mL) was added then the suspension was stirred gently for 16 h. The solid was collected by filtration,

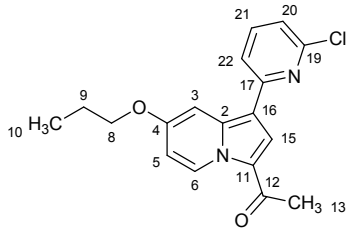
washed with $Et_2O$ then dried under vacuum to yield the product as a white solid (558 mg, 75%); mp 170-174°C; $\nu_{max}$ (neat) 3002 (C-H), 2981 (C-H), 2990 (C-H), 1728 (C=O); $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 0.98 (t, *J*=7.5 Hz, 3 H, C(15)$H_3$), 1.71 - 1.87 (m, 2 H, C(9)$H_2$), 2.27 (s, 3 H, C(13)$H_2$), 4.32 (t, *J*=6.5 Hz, 2 H, C(8)$H_2$), 5.70 (s, 2 H, C(11)$H_2$), 7.68 (d, *J*=7.5 Hz, 2 H, C(3)*H*+C(5)*H*), 8.73 (d, *J*=7.5 Hz, 2 H, C(2)*H*+C(6)*H*); $^{13}$C NMR (101 MHz, DMSO-$d_6$) $\delta$ ppm 10.0 (s, 1 C, *C*(10)), 21.4 (s, 1 C, *C*(9)), 27.0 (s, 1 C, *C*(13)), 66.3 (s, 1 C, *C*(11)), 72.2 (s, 1 C, *C*(8)), 113.2 (s, 2 C, *C*(3)+*C*(5)), 147.2 (s, 2 C, *C*(2)+*C*(6)), 170.2 (s, 1 C, *C*(4)), 200.2 (s, 1 C, *C*(12)); LRMS *m/z* (ESI$^+$) 194 [M$^+$]; HRMS (ESI$^+$) found 194.1183, calculated for $C_{11}H_{16}NO_2^+$ 194.1176; HPLC (System E) $t_r$ 3.8 min (>99%).

## 1-[7-Propoxy-1-(pyridine-2-yl)indolizin-3-yl]ethanone (2)



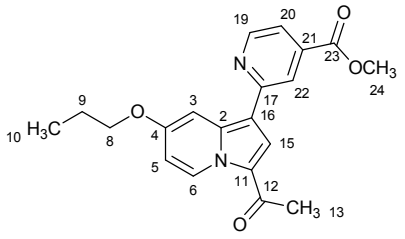

A mixture of compound **4** (84 mg, 0.30 mmol) and $K_2CO_3$ (83 mg, 0.60 mmol) in DMF (2 mL) was stirred for 15 minutes. 2-Ethynylpyridine (36 µL, 0.36 mmol) was added then the resultant mixture was heated at 90 °C for 2 h then allowed to cool. The mixture was partitioned between EtOAc (10 mL) and water (10 mL). The organic phase was washed with water (10 mL) and brine (10 mL) then dried over $MgSO_4$ and evaporated. The crude material was purified by flash column chromatography on a silica column (12 g). The column was eluted with a gradient of EtOAc:*c*-hexane which was increased linearly from 20:80 to 50:50 over 12 CVs. The desired fractions were combined and evaporated to yield the product as a pale brown solid (17 mg, 19%); $R_f$ 0.30 (EtOAc:*c*-hexane, 40:60); mp 125-129 °C; $\nu_{max}$ (neat) 3051 (C-H), 2964 (C-H), 2922 (C-H), 1638 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 1.09 (t, *J*=7.5 Hz, 3 H, C(10)$H_3$), 1.77 - 1.96 (m, 2 H, C(9)$H_2$), 2.57 (s, 3 H, C(13)$H_3$), 4.08 (t, *J*=6.5 Hz, 2 H, C(8)$H_2$), 6.66 (dd, *J*=7.5, 3.0 Hz, 1 H, C(5)*H*), 7.09 (ddd, *J*=7.5, 5.0, 1.0 Hz, 1 H, C(20)*H*), 7.57 - 7.64 (m, 1 H, C(22)*H*), 7.64 - 7.73 (m, 1 H, C(21)*H*), 7.83 (s, 1 H, C(15)*H*), 8.11 (d, *J*=3.0 Hz, 1 H, C(3)*H*), 8.65 (d, *J*=5.0 Hz, 1 H, C(19)*H*), 9.79 (dd, *J*=7.5, 1.0 Hz, 1 H, C(6)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 10.5 (s, 1 C, *C*(10)), 22.3 (s, 1 C, *C*(9)), 26.8 (s, 1 C, *C*(13)), 69.7 (s, 1 C, *C*(8)), 98.5 (s, 1 C, *C*(3)), 108.6 (s, 1 C, *C*(5)), 112.9 (s, 1 C, *C*(16)), 119.8 (s, 1 C, *C*(20/22)), 119.8 (s, 1 C, *C*(20/22)), 121.6 (s, 1 C, *C*(11)), 123.2 (s, 1 C, *C*(15)), 130.1 (s, 1 C, *C*(6)), 136.3 (s, 1 C, *C*(21)), 139.2 (s, 1 C, *C*(2)), 149.2 (s, 1 C, *C*(19)), 154.7 (s, 1 C, *C*(17)), 157.7 (s, 1 C, *C*(4)), 185.6 (s, 1 C, *C*(12)); LRMS *m/z* (ESI$^+$) 295 [MH$^+$]; HRMS (ESI$^+$) found 295.1431, calculated for $C_{18}H_{19}N_2O_2^+$ 295.1441; HPLC (System E) $t_r$ 4.3 min (98%).

## 1-[1-(6-Chloropyridin-2-yl)-7-propoxyindolizin-3-yl]ethanone (16)



A mixture of compound **4** (84 mg, 0.30 mmol) and $K_2CO_3$ (83 mg, 0.60 mmol) in DMF (2 mL) was stirred for 15 minutes. 2-Chloro-6-ethynylpyridine (50 mg, 0.36 mmol) was added then the resultant mixture was heated at 90 °C for 2 h then allowed to cool. The mixture was partitioned between EtOAc (10 mL) and water (10 mL). The organic phase was washed with water (10 mL) and brine (10 mL) then dried over $MgSO_4$ and evaporated. The crude material was purified by flash column chromatography on a silica column (12 g). The column was eluted with a gradient of EtOAc:*c*-hexane which was increased linearly from 20:80 to 50:50 over 12 CVs. The desired fractions were combined and evaporated to yield the product as a beige solid (38 mg, 39%); $R_f$ 0.35 (EtOAc:*c*-hexane, 40:60); mp 184-188 °C; $\nu_{max}$ (neat) 3106 (C-H), 2963 (C-H), 2939 (C-H), 2881 (C-H), 1642 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 1.10 (t, *J*=7.5 Hz, 3 H, C(10)$H_3$), 1.84 - 1.97 (m, 2 H, C(9)$H_2$), 2.55 (s, 3 H, C(13)$H_3$), 4.10 (t, *J*=6.5 Hz, 2 H, C(8)$H_2$), 6.66 (dd, *J*=7.5, 2.5 Hz, 1 H, C(5)*H*), 7.07 (dd, *J*=8.0, 1.0 Hz, 1 H, C(20)*H*), 7.49 (dd, *J*=8.0, 1.0 Hz, 1 H, C(22)*H*), 7.60 (t, *J*=8.0 Hz, 1 H, C(21)*H*), 7.77 (s, 1 H, C(15)*H*), 8.19 (d, *J*=2.5 Hz, 1 H, C(3)*H*), 9.74 (dd, *J*=7.5, 0.5 Hz, 1 H, C(6)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 10.4 (s, 1 C, *C*(10)), 22.1 (s, 1 C, *C*(9)), 26.8 (s, 1 C, *C*(13)), 69.7 (s, 1 C, *C*(8)), 98.8 (s, 1 C, *C*(3)), 108.9 (s, 1 C, *C*(5)), 111.0 (s, 1 C, *C*(16)), 117.4 (s, 1 C, *C*(22)), 119.4 (s, 1 C, *C*(20)), 121.8 (s, 1 C, *C*(11)), 122.9 (s, 1 C, *C*(15)), 130.1 (s, 1 C, *C*(6)), 138.6 (s, 1 C, *C*(21)), 139.5 (s, 1 C, *C*(2)), 150.4 (s, 1 C, *C*(19)), 155.1 (s, 1 C, *C*(17)), 158.0 (s, 1 C, *C*(4)), 185.8 (s, 1 C, *C*(12)); LRMS *m/z* (ESI$^+$) 329 [MH$^+$]; HRMS (ESI$^+$) found 351.0874, calculated for $C_{18}H_{17}ClN_2NaO_2^+$ 351.0871; HPLC (System E) $t_r$ 7.3 min (97%).

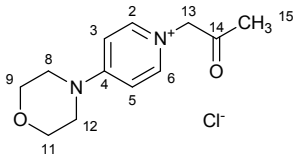
## Methyl 2-(3-acetyl-7-propoxyindolizin-1-yl)pyridine-4-carboxylate (22)



A mixture of compound **4** (84 mg, 0.30 mmol) and $K_2CO_3$ (83 mg, 0.60 mmol) in DMF (2 mL) was stirred for 15 minutes. Methyl 2-ethynylisonicotinate (58 mg, 0.36 mmol) was added then the resultant mixture was heated at 90 °C for 2 h then allowed to cool. The mixture was partitioned between EtOAc (10 mL) and water (10 mL). The organic phase was washed with water (10 mL) and brine (10 mL) then dried over $MgSO_4$ and evaporated. The crude material was purified by flash column chromatography on a silica column (12 g). The column was eluted with a gradient of EtOAc:*c*-hexane which was increased linearly from 20:80 to 50:50 over 12 CVs. The desired fractions were combined and evaporated then the resultant material was purified further by flash column chromatography on a C-18 column (13 g). The column was eluted with a gradient of

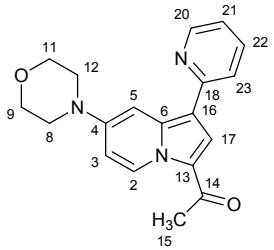
$H_2O$:MeCN (+0.1% $CF_3CO_2H$) which was increased linearly from 95:5 to 95:5 over 30 CVs. The desired fractions were combined and concentrated *in vacuo* to remove most of the solvent. The residual material was partitioned between $CH_2Cl_2$ (5 mL) and saturated aq. $NaHCO_3$ (5 mL). The organic phase was separated then dried over $MgSO_4$ and evaporated to yield the product as a yellow solid (25 mg, 24%); $R_f$ 0.30 (EtOAc:*c*-hexane, 40:60); mp 154-159°C; $\nu_{max}$ (neat) 3088 (C-H), 2976 (C-H), 2942 (C-H), 1727 (ester C=O), 1643 (ketone C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 1.01 (t, *J*=7.5 Hz, 3 H, C(10)$H_3$), 1.68 - 1.93 (m, 2 H, C(9)$H_2$), 2.51 (s, 3 H, C(13)$H_3$), 3.78 - 3.95 (m, 3 H, C(24)$H_3$), 4.00 (t, *J*=6.5 Hz, 2 H, C(8)$H_2$), 6.60 (dd, *J*=7.5, 3.0 Hz, 1 H, C(5)*H*), 7.50 (dd, *J*=5.0, 1.5 Hz, 1 H, C(20)*H*), 7.82 (s, 1 H, C(15)*H*), 8.06 - 8.08 (m, 1 H, C(22)*H*), 8.09 (d, *J*=3.0 Hz, 1 H, C(3)*H*), 8.67 (dd, *J*=5.0, 1.5 Hz, 1 H, C(19)*H*), 9.70 (d, *J*=7.5 Hz, 1 H, C(6)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 10.5 (s, 1 C, *C*(10)), 22.3 (s, 1 C, *C*(9)), 26.9 (s, 1 C, *C*(13)), 52.7 (s, 1 C, *C*(24)), 69.8 (s, 1 C, *C*(8)), 98.8 (s, 1 C, *C*(3)), 108.7 (s, 1 C, *C*(5)), 112.0 (s, 1 C, *C*(16)), 118.2 (s, 1 C, *C*(20)), 118.9 (s, 1 C, *C*(22)), 121.9 (s, 1 C, *C*(11)), 123.3 (s, 1 C, *C*(15)), 130.1 (s, 1 C, *C*(6)), 137.5 (s, 1 C, *C*(21)), 139.4 (s, 1 C, *C*(2)), 149.9 (s, 1 C, *C*(19)), 155.7 (s, 1 C, *C*(17)), 158.0 (s, 1 C, *C*(4)), 166.1 (s, 1 C, *C*(23)), 185.9 (s, 1 C, *C*(12)); LRMS *m/z* (ESI$^+$) 353 [MH$^+$]; HRMS (ESI$^+$) found 375.1309, calculated for $C_{20}H_{20}N_2NaO_4^+$ 375.1315; HPLC (System E) $t_r$ 6.5 min (97%).

### 4-(Morpholin-4-yl)-1-(2-oxopropyl)pyridinium chloride (5)




Chloroacetone (0.820 mL, 10.0 mmol) was added to a stirred solution of 4-morpholinopyridine (1.64 g, 10.0 mmol) in THF (5 mL). The resultant mixture was left to stir for 16 h at room temperature then diluted with EtOAc (5 mL). The solid was filtered then dried under vacuum to yield the product as a white solid (2.15 g, 84%); mp 304-307 °C; $\nu_{max}$ (neat) 3039 (C-H), 3000 (C-H), 2941 (C-H), 2857 (C-H), 1724 (C=O); $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 2.22 (s, 3 H, C(15)$H_3$), 3.64 - 3.79 (m, 8 H, C(8)$H_2$+C(9)$H_2$+C(11)$H_2$+C(12)$H_2$), 5.36 (s, 2 H, C(13)$H_2$), 7.30 (d, *J*=8.0 Hz, 2 H, C(3)*H*+C(5)*H*), 8.17 (d, *J*=8.0 Hz, 2 H, C(2)*H*+C(6)*H*); $^{13}$C NMR (101 MHz, DMSO-$d_6$) $\delta$ ppm 26.9 (s, 1 C, *C*(15)), 46.1 (s, 2 C, *C*(8)+*C*(12)), 64.6 (s, 1 C, *C*(13)), 65.5 (s, 2 C, *C*(9)+*C*(11)), 107.6 (s, 2 C, *C*(3)+*C*(5)), 143.6 (s, 2 C, *C*(2)+*C*(6)), 155.7 (s, 1 C, *C*(4)), 201.0 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 221 [M$^+$]; HRMS (ESI$^+$) found 221.1282, calculated for $C_{12}H_{17}N_2O_2$ 221.1285$^+$; HPLC (System E) $t_r$ 0.4 min (80%).

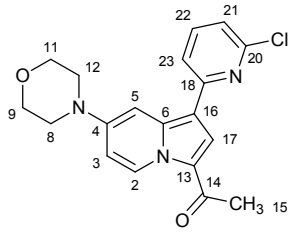
### 1-[7-(Morpholin-4-yl)-1-(pyridin-2-yl)indolizin-3-yl]ethanone (3)




A mixture of compound **5** (80 mg, 0.31 mmol), 2-ethynylpyridine (38 µL, 0.37 mmol) and $K_2CO_3$ (86 mg, 0.62 mmol) in DMF (2 mL) was heated in a sealed vial for 2 h at 90 °C then allowed to cool. The mixture was then partitioned between water (3 mL) and EtOAc (3 mL). The phases were separated then the organic phase was washed with water (3 mL) and brine (3 mL) then dried over $MgSO_4$ and evaporated. The crude material was

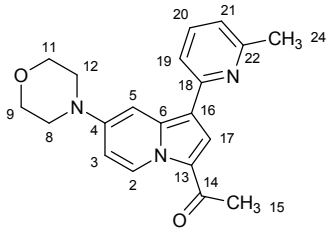
purified by flash column chromatography on a silica column (4 g). The column was eluted with a gradient of EtOAc:*c*-hexane which was increased linearly from 40:60 to 100:0 over 30 CVs. The desired fractions were combined and evaporated to yield the product as a yellow solid (17 mg, 17%); $R_f$ 0.35 (EtOAc) ; mp 180-185 °C; $\nu_{max}$ (neat) 3111 (C-H), 2964 (C-H), 2849 (C-H), 1642 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 2.54 - 2.71 (m, 3 H, C(15)$H_3$), 3.32 - 3.48 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 3.78 - 3.96 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 6.69 (dd, *J*=8.0, 2.5 Hz, 1 H, C(3)*H*), 7.07 (ddd, *J*=7.5, 5.0, 1.0 Hz, 1 H, C(21)*H*), 7.59 - 7.64 (m, 1 H, C(23)*H*), 7.64 - 7.73 (m, 1 H, C(22)*H*), 7.82 (s, 1 H, C(17)*H*), 8.05 (d, *J*=2.5 Hz, 1 H, C(5)*H*), 8.58 - 8.68 (m, 1 H, C(20)*H*), 9.77 (d, *J*=7.8 Hz, 1 H, C(2)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 26.7 (s, 1 C, *C*(15)), 47.6 (s, 2 C, *C*(8)+*C*(12)), 66.5 (s, 2 C, *C*(9)+*C*(11)), 99.5 (s, 1 C, *C*(5)), 106.0 (s, 1 C, *C*(3)), 112.3 (s, 1 C, *C*(16)), 119.6 (s, 1 C, *C*(21/23)), 119.8 (s, 1 C, *C*(21/23)), 121.3 (s, 1 C, *C*(13)), 123.5 (s, 1 C, *C*(17)), 129.7 (s, 1 C, *C*(2)), 136.3 (s, 1 C, *C*(22)), 139.3 (s, 1 C, *C*(6)), 148.7 (s, 1 C, *C*(4)), 149.1 (s, 1 C, *C*(20)), 154.8 (s, 1 C, *C*(18)), 185.1 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 322 [MH$^+$]; HRMS (ESI$^+$) found 322.1538, calculated for $C_{19}H_{20}N_3O_2^+$ 322.1550; HPLC (System E) $t_r$ 3.4 min (97%).

## 1-[1-(6-Chloropyridin-2-yl)-7-(morpholin-4-yl)indolizin-3-yl]ethanone (15)



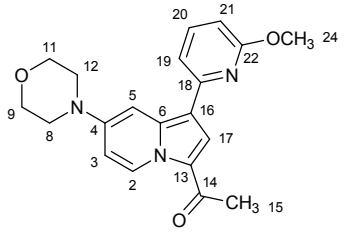

$K_2CO_3$ (323 mg, 2.34 mmol) was added to a solution of compound **5** (300 mg, 1.17 mmol) in DMF (10 mL). The resultant suspension was stirred at room temperature for 15 minutes then 2-ethynylpyridine (189 µL, 1.40 mmol) was added. The mixture was then heated at 90 °C for 2 h then allowed to cool. The solvent was evaporated by $N_2$ blow-down then the mixture was then partitioned between water (5 mL) and $CH_2Cl_2$ (5 mL). The organic phase was collected by passing it through a hydrophobic frit then concentrated by nitrogen blow-down. The crude material was purified by flash column chromatography on a silica column (20 g). The column was eluted with a gradient of EtOAc:*c*-hexane which was increased linearly from 40:60 to 80:20 over 12 CVs. The desired fractions were combined and evaporated to a yellow solid (180 mg). This material was suspended in MeOH (3 mL) then the supernatant was decanted off with a pipette. This process was repeated (×2) then the residual solid was dried under vacuum to yield the product as a yellow solid (166 mg, 40%) $R_f$ 0.45 (EtOAc); mp 215-220 °C; $\nu_{max}$ (neat) 3104 (C-H), 2964 (C-H), 2864 (C-H), 1641 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 2.54 (s, 3 H, C(15)$H_3$), 3.34 - 3.43 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 3.81 - 4.02 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 6.69 (dd, *J*=8.0, 3.0 Hz, 1 H, C(3)*H*), 7.05 (d, *J*=7.5 Hz, 1 H, C(21)*H*), 7.50 (d, *J*=7.5 Hz, 1 H, C(23)*H*), 7.60 (t, *J*=7.5 Hz, 1 H, C(22)*H*), 7.77 (s, 1 H, C(17)*H*), 8.15 (d, *J*=3.0 Hz, 1 H, C(5)*H*), 9.73 (d, *J*=8.0 Hz, 1 H, C(2)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 26.7 (s, 1 C, *C*(15)), 47.4 (s, 2 C, *C*(8)+*C*(12)), 66.5 (s, 2 C, *C*(9)+*C*(11)), 99.7 (s, 1 C, *C*(5)), 106.0 (s, 1 C, *C*(3)), 110.4 (s, 1 C, *C*(16)), 117.3 (s, 1 C, *C*(23)), 119.1 (s, 1 C, *C*(21)), 121.4 (s, 1 C, *C*(13)), 123.2 (s, 1 C, *C*(17)), 129.7 (s, 1 C, *C*(2)), 138.6 (s, 1 C, *C*(22)), 139.6 (s, 1 C, *C*(6)), 148.9 (s, 1 C, *C*(4)), 150.4 (s, 1 C, *C*(20)), 155.3 (s, 1 C, *C*(18)), 185.2 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 356 [MH$^+$]; HRMS (ESI$^+$) found 378.0979, calculated for $C_{19}H_{18}ClN_3NaO_2^+$ 378.0980; HPLC (System D) $t_r$ 17.8 min (91%).

**1-[1-(6-Methylpyridin-2-yl)-7-(morpholin-4-yl)indolizin-3-yl]ethanone (17)**



$K_2CO_3$ (47 mg, 0.34 mmol) was added to a solution of compound **5** (80 mg, 0.31 mmol) in DMF (2 mL). The resultant suspension was stirred at room temperature for 10 minutes then 2-ethynyl-6-methylpyridine (40 mg, 0.34 mmol) was added. The mixture was then heated at 90 °C for 2 h then allowed to cool. The mixture was then partitioned between water (3 mL) and $CH_2Cl_2$ (3 mL). The organic phase was collected by passing it through a hydrophobic frit then concentrated by nitrogen blow-down. The crude material was purified by flash column chromatography on a silica column (4 g). The column was eluted with a gradient of EtOAc:*c*-hexane + 0.1% $NEt_3$ which was increased linearly from 40:60 to 80:20 over 20 CVs. The desired fractions were combined and evaporated. The resultant solid was suspended in MeOH then filtered to yield the product as a yellow solid (29 mg, 28%); $R_f$ 0.25 (EtOAc:*c*-hexane, 60:40 + 1% $NEt_3$); mp 168-173 °C; $\nu_{max}$ (neat) 3113 (C-H), 2950 (C-H), 2849 (C-H), 1640 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 2.55 (s, 3 H, C(15)$H_3$), 2.62 (s, 3 H, C(24)$H_3$), 3.31 - 3.50 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 3.79 - 3.96 (m, 4 H, C(9)$H_2$, C(11)$H_2$), 6.69 (dd, *J*=8.0, 2.5 Hz, 1 H, C(3)*H*), 6.95 (d, *J*=7.5 Hz, 1 H, C(21)*H*), 7.43 (d, *J*=7.5 Hz, 1 H, C(19)*H*), 7.59 (t, *J*=7.5 Hz, 1 H, C(20)*H*), 7.83 (s, 1 H, C(17)*H*), 8.14 (s, 1 H, C(5)*H*), 9.76 (d, *J*=8.0 Hz, 1 H, C(2)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 24.7 (s, 1 C, *C*(24)), 26.7 (s, 1 C, *C*(15)), 47.6 (s, 2 C, *C*(8)+*C*(12)), 66.5 (s, 2 C, *C*(9)+*C*(11)), 99.9 (s, 1 C, *C*(5)), 106.1 (s, 2 C, *C*(3)+*C*(16)), 116.8 (s, 1 C, *C*(19)), 119.0 (s, 1 C, *C*(21)), 121.2 (s, 1 C, *C*(13)), 123.5 (s, 1 C, *C*(17)), 129.7 (s, 1 C, *C*(2)), 136.7 (s, 1 C, *C*(20)), 139.3 (s, 1 C, *C*(6)), 148.6 (s, 1 C, *C*(4)), 154.0 (s, 1 C, *C*(18/22)), 157.3 (s, 1 C, *C*(18/22)), 185.1 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 336 [MH$^+$]; HRMS (ESI$^+$) found 336.1692, calculated for $C_{20}H_{22}N_3O_2^+$ 336.1707; HPLC (System D) $t_r$ 9.5 min (97%).

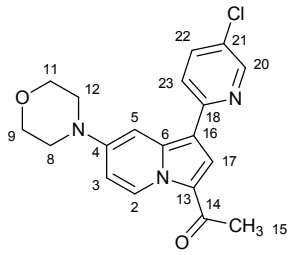
**1-[1-(6-Methoxypyridin-2-yl)-7-(morpholin-4-yl)indolizin-3-yl]ethanone (18)**



$K_2CO_3$ (47 mg, 0.34 mmol) was added to a solution of compound **5** (80 mg, 0.31 mmol) in DMF (2 mL). The resultant suspension was stirred at room temperature for 10 minutes then 2-ethynyl-6-methoxypyridine (46 mg, 0.34 mmol) was added. The mixture was then heated at 90 °C for 2 h then allowed to cool. The mixture was then partitioned between water (3 mL) and $CH_2Cl_2$ (3 mL). The organic phase was collected by passing it through a hydrophobic frit then concentrated by nitrogen blow-down. The crude material was purified by flash column chromatography on a silica column (4 g). The column was eluted with a gradient of EtOAc:*c*-hexane + 0.1% $NEt_3$ which was increased linearly from 40:60 to 80:20 over 20 CVs. The desired fractions were combined and evaporated. The resultant solid was suspended in MeOH then filtered to yield the

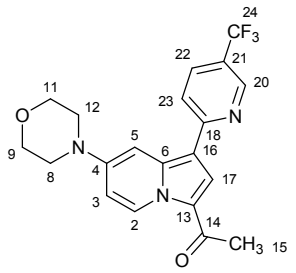
product as a yellow solid (29 mg, 28%); $R_f$ 0.25 (EtOAc:*c*-hexane, 60:40 + 1% $NEt_3$); mp 177-181 °C; $\nu_{max}$ (neat) 3124 (C-H), 2957 (C-H), 2821 (C-H), 1643 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 2.55 (s, 3 H, C(15)$H_3$), 3.21 - 3.36 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 3.84 - 3.97 (m, 4 H, C(9)$H_2$+C(12)$H_2$), 4.07 (s, 3 H, C(24)$H_3$), 6.55 (d, *J*=8.0 Hz, 1 H, C(21)*H*), 6.68 (dd, *J*=8.0, 3.0 Hz, 1 H, C(3)*H*), 7.23 (d, *J*=8.0 Hz, 1 H, C(19)*H*), 7.58 (t, *J*=8.0 Hz, 1 H, C(20)*H*), 7.79 (s, 1 H, C(17)*H*), 8.15 (d, *J*=3.0 Hz, 1 H, C(5)*H*), 9.75 (d, *J*=8.0 Hz, 1 H, C(2)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 26.7 (s, 1 C, *C*(15)), 47.9 (s, 2 C, *C*(8)+*C*(12)), 53.2 (s, 1 C, *C*(24)), 66.4 (s, 2 C, *C*(9)+*C*(11)), 100.2 (s, 1 C, *C*(5)), 106.3 (s, 2 C, *C*(3)+*C*(21)), 112.1 (s, 1 C, *C*(19)), 112.3 (s, 1 C, *C*(16)), 121.2 (s, 1 C, *C*(13)), 123.4 (s, 1 C, *C*(17)), 129.7 (s, 1 C, *C*(2)), 138.8 (s, 1 C, *C*(20)), 139.1 (s, 1 C, *C*(6)), 148.4 (s, 1 C, *C*(4)), 152.4 (s, 1 C, *C*(18)), 163.4 (s, 1 C, *C*(22)), 185.2 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 352 [MH$^+$]; HRMS (ESI$^+$) found 352.1665, calculated for $C_{20}H_{22}N_3O_2^+$ 352.1656; HPLC (System D) $t_r$ 17.0 min (94%).

**1-[1-(5-Chloropyridin-2-yl)-7-(morpholin-4-yl)indolizin-3-yl]ethanone (19)**



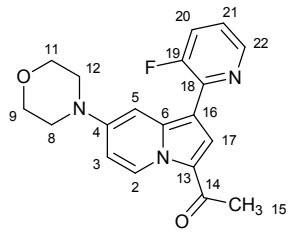

$K_2CO_3$ (108 mg, 0.78 mmol) was added to a solution of compound **5** (100 mg, 0.39 mmol) in DMF (2 mL). The resultant suspension was stirred at room temperature for 15 minutes then 5-chloro-2-ethynylpyridine (59 mg, 0.43 mmol) was added. The mixture was then heated at 90 °C for 2 h then allowed to cool. The solvent was evaporated by $N_2$ blow-down then the mixture was then partitioned between water (5 mL) and $CH_2Cl_2$ (5 mL). The organic phase was collected by passing it through a hydrophobic frit then concentrated by nitrogen blow-down. The crude material was purified by flash column chromatography on a silica column (12 g). The column was eluted with a gradient of EtOAc:*c*-hexane + 0.1% $NEt_3$ which was increased linearly from 40:60 to 80:20 over 12 CVs. The desired fractions were combined and evaporated to an orange solid (39 mg). This material was suspended in MeOH, refluxed, allowed to cool, then filtered to yield the product as a yellow solid (34 mg, 25%); $R_f$ 0.30 (EtOAc:*c*-hexane, 60:40 + 1% $NEt_3$); mp 282-286 °C; $\nu_{max}$ (neat) 3114 (C-H), 2958 (C-H), 2923 (C-H), 2870 (C-H), 1642 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 2.54 (s, 3 H, C(15)$H_3$), 3.29 - 3.39 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 3.83 - 3.95 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 6.70 (dd, *J*=7.8, 2.7 Hz, 1 H, C(3)*H*), 7.55 (d, *J*=8.5 Hz, 1 H, C(23)*H*), 7.64 (dd, *J*=8.5, 2.5 Hz, 1 H, C(22)*H*), 7.76 (s, 1 H, C(17)*H*), 7.93 - 8.01 (m, 1 H, C(5)*H*), 8.56 (d, *J*=2.0 Hz, 1 H, C(20)*H*), 9.69 - 9.78 (m, 1 H, C(2)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 26.5 (s, 1 C, *C*(15)), 47.4 (s, 2 C, *C*(8)+*C*(12)), 66.4 (s, 2 C, *C*(9)+*C*(11)), 99.3 (s, 1 C, *C*(5)), 106.0 (s, 1 C, *C*(3)), 111.3 (s, 1 C, *C*(16)), 120.4 (s, 1 C, *C*(23)), 121.2 (s, 1 C, *C*(13)), 123.6 (s, 1 C, *C*(17)), 127.3 (s, 1 C, *C*(21)), 129.7 (s, 1 C, *C*(2)), 136.0 (s, 1 C, *C*(22)), 139.4 (s, 1 C, *C*(6)), 147.7 (s, 1 C, *C*(20)), 148.9 (s, 1 C, *C*(4)), 152.9 (s, 1 C, *C*(18)), 185.2 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 356 [MH$^+$]; HRMS (ESI$^+$) found 378.0971, calculated for $C_{19}H_{18}ClN_3NaO_2^+$ 378.0980; HPLC (System E) $t_r$ 5.9 min (96%).

**1-{7-(Morpholin-4-yl)-1-[5-(trifluoromethyl)pyridin-2-yl]indolizin-3-yl}ethanone (20)**



$K_2CO_3$ (108 mg, 0.78 mmol) was added to a solution of compound **5** (100 mg, 0.39 mmol) in DMF (2 mL). The resultant suspension was stirred at room temperature for 15 minutes then 5-2-ethynyl-5-(trifluoromethyl)pyridine (74 mg, 0.43 mmol) was added. The mixture was then heated at 90 °C for 2 h then allowed to cool. The solvent was evaporated by $N_2$ blow-down then the mixture was then partitioned between water (5 mL) and $CH_2Cl_2$ (5 mL). The organic phase was collected by passing it through a hydrophobic frit then concentrated by nitrogen blow-down. The crude material was purified by flash column chromatography on a silica column (12 g). The column was eluted with a gradient of EtOAc:*c*-hexane + 0.1% $NEt_3$ which was increased linearly from 40:60 to 80:20 over 12 CVs. The desired fractions were combined and evaporated to a yellow solid (40 mg). The material thus obtained was purified further by flash column chromatography on a C-18 column (13 g). The column was eluted with a gradient of $H_2O$:MeCN (+0.1% $CF_3CO_2H$) which was increased linearly from 95:5 to 5:95 over 20 CVs. The desired fractions were combined and evaporated then partitioned between $CH_3Cl$ (5 mL) and saturated aq. $NaHCO_3$ (5 mL). The organic phase was collected by passing through a hydrophobic frit then evaporated to yield the product as a yellow solid (26 mg, 17%); $R_f$ 0.30 (EtOAc:*c*-hexane, 60:40 + 1% $NEt_3$); mp 271-278 °C; $\nu_{max}$ (neat) 3102 (C-H), 2966 (C-H), 2854 (C-H), 1644 (C=O); $^1$H NMR (200 MHz, $CDCl_3$) $\delta$ ppm 2.58 (s, 3 H, C(15)$H_3$), 3.35 - 3.48 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 3.87 - 3.98 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 6.74 (dd, *J*=8.0, 3.0 Hz, 1 H, C(3)*H*), 7.71 (m, 1 H, C(23)*H*), 7.83 - 7.92 (m, 2 H, C(17)*H*+C(22)*H*), 8.17 (d, *J*=3.0 Hz, 1 H, C(5)*H*), 8.87 - 8.95 (m, 1 H, C(20)*H*), 9.79 (d, *J*=8.0 Hz, 1 H, C(2)*H*); $^{13}$C NMR (126 MHz, CHLOROFORM-*d*) $\delta$ ppm 26.8 (s, 1 C, *C*(15)), 47.4 (s, 2 C, *C*(8)+*C*(12)), 66.4 (s, 2 C, *C*(9)+*C*(11)), 99.6 (s, 1 C, *C*(5)), 106.0 (s, 1 C, *C*(3)), 110.3 - 110.5 (m, 1 C, *C*(16)), 118.8 (s, 1 C, *C*(23)), 124.0 (q, *J*=271.5 Hz, 1 C, *C*(24)), 121.5 (q, *J*=33.0 Hz, 1 C, *C*(21)), 121.8 (s, 1 C, *C*(13)), 123.9 (s, 1 C, *C*(17)), 129.9 (s, 1 C, *C*(2)), 132.9 - 133.3 (m, 1 C, *C*(22)), 139.9 (s, 1 C, *C*(6)), 145.9 - 146.1 (m, 1 C, *C*(20)), 149.3 (s, 1 C, *C*(4)), 157.7 - 158.1 (m, 1 C, *C*(18)), 185.6 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 390 [MH$^+$]; HRMS (ESI$^+$) found 412.1259, calculated for $C_{20}H_{18}F_3N_3NaO_3^+$ 412.1243; HPLC (System D) $t_r$ 18.1 min (91%).

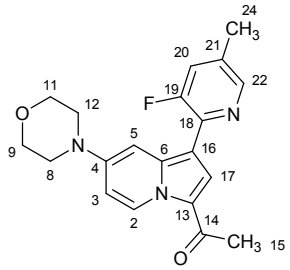
**1-[1-(3-Fluoropyridin-2-yl)-7-(morpholin-4-yl)indolizin-3-yl]ethanone (24)**



$K_2CO_3$ (47 mg, 0.34 mmol) was added to a solution of compound **5** (80 mg, 0.31 mmol) in DMF (2 mL). The resultant suspension was stirred at room temperature for 10 minutes then 2-ethynyl-3-fluoropyridine (42

mg, 0.34 mmol) was added. The mixture was then heated at 90 °C for 2 h then allowed to cool. The mixture was then partitioned between water (3 mL) and $CH_2Cl_2$ (3 mL). The organic phase was collected by passing it through a hydrophobic frit then concentrated by nitrogen blow-down. The crude material was purified by flash column chromatography on a silica column (4 g). The column was eluted with a gradient of EtOAc:*c*-hexane + 0.1% $NEt_3$ which was increased linearly from 40:60 to 80:20 over 20 CVs. The desired fractions were combined and evaporated. The resultant solid was suspended in MeOH then filtered to yield the product as a yellow solid (16 mg, 28%); $R_f$ 0.25 (EtOAc:*c*-hexane, 60:40 + 1% $NEt_3$); mp 191-195 °C; $\nu_{max}$ (neat) 3104 (C-H), 2961 (C-H), 2925 (C-H), 2864 (C-H), 1645 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 2.56 (s, 3 H, C(15)$H_3$), 3.22 - 3.39 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 3.82 - 4.03 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 6.72 (dd, *J*=8.0, 3.0 Hz, 1 H, C(3)*H*), 7.07 - 7.24 (m, 1 H, C(21)*H*), 7.44 (ddd, *J*=12.0, 8.0, 1.5 Hz, 1 H, C(20)*H*), 8.03 (d, *J*=3.0 Hz, 1 H, C(17)*H*), 8.12 (d, *J*=3.0 Hz, 1 H, C(5)*H*), 8.46 (dt, *J*=4.5, 1.5 Hz, 1 H, C(22)*H*), 9.80 (d, *J*=8.0 Hz, 1 H, C(2)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 26.7 (s, 1 C, *C*(15)), 47.6 (s, 2 C, *C*(8)+*C*(12)), 66.5 (s, 2 C, *C*(9)+*C*(11)), 99.7 (s, 1 C, *C*(5)), 106.1 (s, 1 C, *C*(3)), 106.9 (d, *J*=5.5 Hz, 1 C, *C*(3)), 120.1 (d, *J*=4.0 Hz, 1 C, *C*(21)), 121.4 (d, *J*=3.0 Hz, 1 C, *C*(13)), 122.9 (d, *J*=20.0 Hz, 1 C, *C*(20)), 126.0 (d, *J*=15.0 Hz, 1 C, *C*(17)), 129.7 (s, 1 C, *C*(2)), 140.1 (s, 1 C, *C*(6)), 143.7 (d, *J*=10.5 Hz, 1 C, *C*(18)), 144.5 (d, *J*=5.0 Hz, 1 C, *C*(22)), 148.8 (s, 1 C, *C*(4)), 156.3 (d, *J*=259.0 Hz, 1 C, *C*(19)), 185.5 (s, 1 C, *C*(14)); $^{19}$F NMR (377 MHz, $CDCl_3$) $\delta$ ppm -121.8 (s, 1 F); LRMS *m/z* ($ESI^+$) 340 [$MH^+$]; HRMS ($ESI^+$) found 340.1440, calculated for $C_{19}H_{19}FN_3O_2^+$ 340.1456; HPLC (System D) $t_r$ 15.3 min (97%).

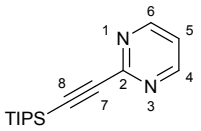
**1-[1-(3-Fluoro-5-methylpyridin-2-yl)-7-(morpholin-4-yl)indolizin-3-yl]ethanone (25)**




$K_2CO_3$ (47 mg, 0.34 mmol) was added to a solution of compound **5** (80 mg, 0.31 mmol) in DMF (2 mL). The resultant suspension was stirred at room temperature for 10 minutes then 2-ethynyl-3-fluoro-5-methylpyridine (46 mg, 0.34 mmol) was added. The mixture was then heated at 90 °C for 2 h then allowed to cool. The mixture was then partitioned between water (3 mL) and $CH_2Cl_2$ (3 mL). The organic phase was collected by passing it through a hydrophobic frit then concentrated by nitrogen blow-down. The crude material was purified by flash column chromatography on a silica column (4 g). The column was eluted with a gradient of EtOAc:*c*-hexane + 0.1% $NEt_3$ which was increased linearly from 40:60 to 80:20 over 20 CVs. The desired fractions were combined and evaporated. The resultant solid was suspended in MeOH then filtered to yield the product as a yellow solid (16 mg, 28%); $R_f$ 0.25 (EtOAc:*c*-hexane, 60:40 + 1% $NEt_3$); mp 235-239 °C; $\nu_{max}$ (neat) 3119 (C-H), 2965 (C-H), 2917 (C-H), 2861 (C-H), 1643 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 2.31 (s, 3 H, C(24)$H_3$), 2.42 - 2.53 (m, 3 H, C(15)$H_3$), 3.14 - 3.30 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 3.70 - 3.94 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 6.63 (dd, *J*=8.0, 2.5 Hz, 1 H, C(3)*H*), 7.15 - 7.38 (m, 1 H, C(20)*H*), 7.90 (d, *J*=3.0 Hz, 1 H, C(17)*H*), 7.96 (d, *J*=2.5 Hz, 1 H, C(5)*H*), 8.12 - 8.34 (m, 1 H, C(22)*H*), 9.72 (d, *J*=8.0 Hz, 1 H, C(2)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 17.8 (s, 1 C, *C*(24)), 26.7 (s, 1 C, *C*(15)), 47.6 (s, 2 C, *C*(8)+*C*(12)), 66.5 (s, 2 C, *C*(9)+*C*(11)), 99.6 (s, 1 C, *C*(5)), 106.1 (s, 1 C, *C*(3)), 107.1 (d, *J*=6.5 Hz, 1 C, *C*(16)), 121.3 (d, *J*=3.0 Hz, 1 C, *C*(13)), 123.6 (d, *J*=20.0 Hz, 1 C, *C*(20)), 125.7 (d, *J*=13.5 Hz, 1 C, *C*(17)), 129.6 (s, 1 C, *C*(2)), 130.7 (d, *J*=4.0 Hz, 1 C, *C*(2)), 139.8 (s, 1 C, *C*(6)), 140.7 (d, *J*=11.0 Hz, 1 C, *C*(18)), 144.8 (d, *J*=4.0 Hz, 1 C, *C*(22)), 148.6 (s, 1 C, *C*(4)), 156.1 (d, *J*=259.0 Hz, 1 C, *C*(19)), 185.4 (s, 1 C, *C*(14)); $^{19}$F NMR (377 MHz, $CDCl_3$) $\delta$ ppm -123.6 (s, 1 F);

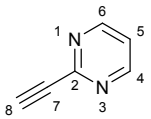
LRMS *m/z* (ESI$^+$) 354 [MH$^+$]; HRMS (ESI$^+$) found 354.1596, calculated for $C_{20}H_{21}FN_3O_2^+$ 354.1612; HPLC (System D) $t_r$ 15.8 min (92%).

## 2-[(Tripropan-2-ylsilyl)ethynyl]pyrimidine



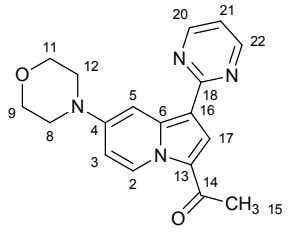

A mixture of 2-bromo-pyrimidine (690 mg, 5.00 mmol), (triisopropylsilyl)acetylene (1.94 mL, 10.0 mmol), $Pd(PPh_3)_2Cl_2$ (152 mg, 0.05 mmol) CuI (10 mg, 0.10 mmol) and $Et_3N$ (1.80 mL, 15.0 mmol) in DMF (20 mL) was degassed by evacuating the apparatus then refilling with nitrogen (×3). The reaction mixture was stirred for 16 h at room temperature then concentrated *in vacuo*. The residue was suspended in EtOAc then pre-adsorbed onto silica. The crude material was purified by flash column chromatography on a silica column (40 g). The column was eluted with a gradient of EtOAc:*c*-hexane which was increased linearly from 0:100 to 40:60 over 12 CVs. The desired fractions were combined and evaporated to yield the product as a brown oil (584 mg, 45%); $R_f$ 0.20 (EtOAc); $\nu_{max}$ (neat) 2944 (C-H), 2892 (C-H), 2866 (C-H), ; $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 1.04 - 1.27 (m, 21 H, (21×TIPS-*H*)), 7.23 (t, *J*=5.0 Hz, 1 H, C(5)*H*), 8.72 (d, *J*=5.0 Hz, 2 H, C(4)*H*+C(6)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 11.2 (s, 3 C, (3×TIPS *C*H)), 18.6 (s, 6C (6×TIPS *C*$H_3$) 91.2 (s, 1 C, *C*(8)), 104.7 (s, 1 C, *C*(7)), 119.9 (s, 1 C, *C*(5)), 152.7 (s, 1 C, *C*(2)), 157.2 (s, 2 C, *C*(4)+*C*(6)); LRMS *m/z* (ESI$^+$) 283 [(M+Na)$^+$], 261 [MH$^+$]; HRMS (ESI$^+$) found 261.1785, calculated for $C_{15}H_{25}N_2Si^+$ 261.1782; HPLC (System D) $t_r$ 17.8 min (92%).

## 2-Ethynylpyrimidine[1]



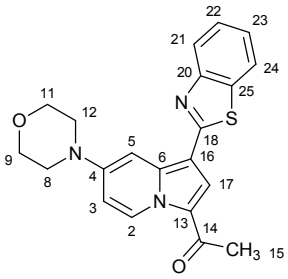

A mixture of 2-[(tripropan-2-ylsilyl)ethynyl]pyrimidine (260 mg, 1.0 mmol) and TBAF on silica (1.5 mmol/g, 733 mg, 1.1 mmol) in THF (10 mL) was stirred at room temperature for 2 h then evaporated. The crude material was purified by flash column chromatography on a silica column (24 g). The column was eluted with a gradient of EtOAc:*c*-hexane which was increased linearly from 20:80 to 60:40 over 12 CVs. The desired fractions were combined and evaporated to yield the product as a pale-brown oil (47 mg, 45%); $R_f$ 0.25 (EtOAc:*c*-hexane, 40:60); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 3.10 (s, 1 H, C(8)*H*), 7.24 (t, *J*=5.0 Hz, 1 H, C(5)*H*), 8.67 (d, *J*=5.0 Hz, 2 H, C(4)*H*+C(6)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 75.8 (s, 1 C, *C*(8)), 81.7 (s, 1 C, *C*(7)), 120.4 (s, 1 C, *C*(5)), 152.1 (s, 1 C, *C*(2)), 157.2 (s, 2 C, *C*(4)+*C*(6)).

## 1-[7-(Morpholin-4-yl)-1-(pyrimidin-2-yl)indolizin-3-yl]ethanone (26)



A mixture of compound **5** (100 mg, 0.39 mmol) and $K_2CO_3$ (108 mg, 0.78 mmol) in DMF (2 mL) was stirred at room temperature for 15 minutes then 2-ethynylpyrimidine (45 mg, 0.43 mmol) was added. The resultant mixture was stirred for 2 h at 90 °C then allowed to cool. The mixture was then partitioned between water (5 mL) and $CH_2Cl_2$ (5 mL). The phases were separated using a hydrophobic frit then the organic phase was evaporated. The crude material was purified by flash column chromatography on a silica column (12 g). The column was eluted with a gradient of EtOAc:*c*-hexane which was increased linearly from 60:40 to 100:0 over 10 CVs. The desired fractions were combined and evaporated to a yellow solid (28 mg). This material thus obtained was suspended in MeOH (5 mL), refluxed, allowed to cool, sonicated, then filtered and dried under vacuum to yield the product as a pale-yellow solid (19 mg, 15%); $R_f$ 0.30 (EtOAc + 1% $NEt_3$); mp 213-217 °C; $\nu_{max}$ (neat) 3114 (C-H), 3023 (C-H), 2858 (C-H), 2831 (C-H), 1643 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 2.50 (s, 3 H, C(15)$H_3$), 3.27 - 3.38 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 3.72 - 3.88 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 6.63 (dd, *J*=8.0, 3.0 Hz, 1 H, C(3)*H*), 6.91 (t, *J*=5.0 Hz, 1 H, C(21)*H*), 8.09 (d, *J*=3.0 Hz, 1 H, C(5)*H*), 8.20 (s, 1 H, C(17)*H*), 8.63 (d, *J*=5.0 Hz, 2 H, C(20)*H*+C(22)*H*), 9.69 (d, *J*=8.0 Hz, 1 H, C(2)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 26.8 (s, 1 C, *C*(15)), 47.5 (s, 2 C, *C*(8)+*C*(12)), 66.5 (s, 2 C, *C*(9)+*C*(11)), 99.9 (s, 1 C, *C*(5)), 105.7 (s, 1 C, *C*(3)), 111.2 (s, 1 C, *C*(16)), 116.1 (s, 1 C, *C*(21)), 121.7 (s, 1 C, *C*(13)), 126.2 (s, 1 C, *C*(17)), 130.0 (s, 1 C, *C*(2)), 140.6 (s, 1 C, *C*(6)), 149.1 (s, 1 C, *C*(4)), 156.8 (s, 2 C, *C*(20)+*C*(22)), 163.1 (s, 1 C, *C*(18)), 186.1 (s, 1 C, *C*(14)); LRMS *m/z* ($ESI^+$) 323 [$MH^+$]; HRMS ($ESI^+$) found 345.1327, calculated for $C_{18}H_{18}N_4NaO_2^+$ 345.1322; HPLC (System E) $t_r$ 4.5 min (96%).

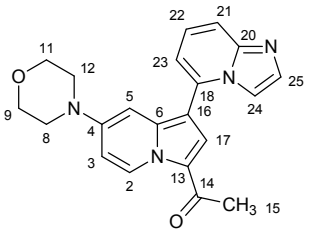
## 1-[1-(1,3-Benzothiazol-2-yl)-7-(morpholin-4-yl)indolizin-3-yl]ethanone (27)



$K_2CO_3$ (47 mg, 0.34 mmol) was added to a solution of compound **5** (80 mg, 0.31 mmol) in DMF (2 mL). The resultant suspension was stirred at room temperature for 10 minutes then 2-ethynyl-1,3-benzothiazole (55 mg, 0.34 mmol) was added. The mixture was then heated at 90 °C for 2 h then allowed to cool. The mixture was then partitioned between water (3 mL) and $CH_2Cl_2$ (3 mL). The organic phase was collected by passing it through a hydrophobic frit then concentrated by nitrogen blow-down. The crude material was purified by flash column chromatography on a silica column (4 g). The column was eluted with a gradient of EtOAc:*c*-hexane + 0.1% $NEt_3$ which was increased linearly from 40:60 to 80:20 over 20 CVs. The desired fractions were combined and evaporated. The resultant solid was suspended in MeOH then filtered to yield the

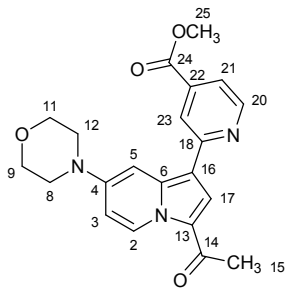
product as an orange solid (14 mg, 12%); $R_f$ 0.25 (EtOAc:*c*-hexane, 60:40 + 1% $NEt_3$); mp 212-217 °C; $\nu_{max}$ (neat) 3061 (C-H), 2963 (C-H), 2856 (C-H), 1645 (C=O); $^1$H NMR (500 MHz, $CDCl_3$) $\delta$ ppm 2.57 (s, 3 H, C(15)$H_3$), 3.39 - 3.49 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 3.84 - 4.01 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 6.72 (dd, *J*=7.5, 2.5 Hz, 1 H, C(3)*H*), 7.32 (t, *J*=7.5 Hz, 1 H, C(22/23)*H*), 7.45 (t, *J*=7.5 Hz, 1 H, C(23/24)*H*), 7.80 - 7.91 (m, 2 H, C(17)*H*+C(21/24)*H*), 7.92 - 8.06 (m, 2 H, C(5)*H*+C(21/24)*H*), 9.73 (d, *J*=8.0 Hz, 1 H, C(2)*H*); $^{13}$C NMR (126 MHz, $CDCl_3$) $\delta$ ppm 26.8 (s, 1 C, *C*(15)), 47.2 (s, 2 C, *C*(8)+*C*(12)), 66.4 (s, 2 C, *C*(9)+*C*(11)), 98.8 (s, 1 C, *C*(5)), 106.0 (s, 1 C, *C*(3)), 107.0 (s, 1 C, *C*(16)), 121.2 (s, 1 C, *C*(21/24)), 121.8 (s, 1 C, *C*(13/21/24)), 121.9 (s, 1 C, *C*(13/21/24)), 124.0 (s, 1 C, *C*(22/23)), 124.9 (s, 1 C, *C*(17)), 126.0 (s, 1 C, *C*(22/23)), 130.1 (s, 1 C, *C*(2)), 133.3 (s, 1 C, *C*(25)), 139.2 (s, 1 C, *C*(6)), 149.5 (s, 1 C, *C*(4)), 154.2 (s, 1 C, *C*(20)), 162.7 (s, 1 C, *C*(18)), 185.8 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 378 [MH$^+$]; HRMS (ESI$^+$) found 378.1274, calculated for $C_{21}H_{20}N_3O_2S^+$ 378.1271; HPLC (System D) $t_r$ 18.4 min (92%).

### 1-[1-(Imidazo[1,2-*a*]pyridin-5-yl)-7-(morpholin-4-yl)indolizin-3-yl]ethanone (28)



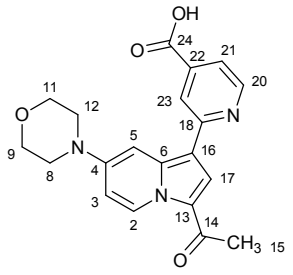

$K_2CO_3$ (108 mg, 0.78 mmol) was added to a solution of compound **5** (100 mg, 0.39 mmol) in DMF (2 mL). The resultant suspension was stirred at room temperature for 15 minutes then 5-ethynylimidazo[1,2*a*]pyridine (61 mg, 0.43 mmol) was added. The mixture was then heated at 90 °C for 2 h then allowed to cool. The solvent was evaporated by $N_2$ blow-down then the mixture was then partitioned between water (5 mL) and $CH_2Cl_2$ (5 mL). The organic phase was collected by passing it through a hydrophobic frit then concentrated by nitrogen blow-down. The crude material was purified by flash column chromatography on a silica column (12 g). The column was eluted with a gradient of EtOAc:MeOH:$NEt_3$ which was increased linearly from 99:1:0.1 to 90:10:1 over 12 CVs. The desired fractions were combined and evaporated to a cream solid (32 mg). The material thus obtained was purified further by flash column chromatography on a C-18 column (4.3 g). The column was eluted with a gradient of $H_2O$:MeCN (+0.1% $CF_3CO_2H$) which was increased linearly from 95:5 to 5:95 over 12 CVs. The desired fractions were combined and evaporated then partitioned between $CH_2Cl_2$ (2 mL) and saturated aq. $NaHCO_3$ (2 mL). The organic phase was collected by passing through a hydrophobic frit then evaporated to yield the product as a yellow solid (5 mg, 4%); $R_f$ 0.25 (EtOAc:MeOH:$NEt_3$, 90:10:1); mp 212-217 °C; $\nu_{max}$ (neat) 3079 (C-H), 2952 (C-H), 2867 (C-H), 1644 (C=O); $^1$H NMR (200 MHz, $CDCl_3$) $\delta$ ppm 2.56 (s, 3 H, C(15)$H_3$), 3.11 - 3.32 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 3.75 - 3.91 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 6.46 (d, *J*=2.4 Hz, 1 H, C(24/25)*H*), 6.75 (dd, *J*=7.9, 2.7 Hz, 1 H, C(3)*H*), 6.88 (dd, *J*=7.0, 1.1 Hz, 1 H, C(23)*H*), 7.23 - 7.35 (m, 1 H, C(22)*H*), 7.54 - 7.74 (m, 4 H, C(5)*H*+C(17)*H*+C(21)*H*+C(24/25)*H*), 9.81 (d, *J*=7.9 Hz, 1 H, C(2)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 26.7 (s, 1 C, *C*(15)), 47.4 (s, 2 C, *C*(8)+*C*(12)), 66.2 (s, 2 C, *C*(9)+*C*(11)), 96.4 (s, 1 C, *C*(5)), 105.8 (s, 1 C, *C*(16)), 106.2 (s, 1 C, *C*(3)), 111.5 (s, 1 C, *C*(24)), 113.0 (s, 1 C, *C*(23)), 115.6 (s, 1 C, *C*(21)), 121.5 (s, 1 C, *C*(13)), 124.4 (s, 1 C, *C*(17)), 124.7 (s, 1 C, *C*(22)), 130.1 (s, 1 C, *C*(2)), 132.8 (s, 1 C, *C*(18)), 132.9 (s, 1 C, *C*(25)), 138.3 (s, 1 C, *C*(6)), 146.1 (s, 1 C, *C*(20)), 148.2 (s, 1 C, *C*(4)), 185.5 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 361 [MH$^+$]; HRMS (ESI$^+$) found 361.1643, calculated for $C_{21}H_{21}N_4O_2^+$ 361.1659; HPLC (System E) $t_r$ 3.8 min (99%).

## Methyl 2-[3-acetyl-7-(morpholin-4-yl)indolizin-1-yl]pyridine-4-carboxylate (21)



$K_2CO_3$ (216 mg, 1.56 mmol) was added to a solution of compound **5** (200 mg, 0.78 mmol) in DMF (4 mL). The resultant suspension was stirred at room temperature for 15 minutes then methyl 2-ethynylisonicotinate (151 mg, 0.93 mmol) was added. The mixture was then heated at 90 °C for 2 h then allowed to cool. The solvent was evaporated by $N_2$ blow-down then the mixture was then partitioned between water (5 mL) and $CH_2Cl_2$ (5 mL). The organic phase was collected by passing it through a hydrophobic frit then concentrated by nitrogen blow-down. The crude material was purified by flash column chromatography on a silica column (12 g). The column was eluted with a gradient of EtOAc:*c*-hexane which was increased linearly from 60:40 to 100:0 over 12 CVs. The desired fractions were combined and evaporated to an orange solid (127 mg). This material was suspended in MeOH (3 mL) then the supernatant was decanted off with a pipette. This process was repeated (×2) then the residual solid was dried under vacuum to yield the product as a yellow solid (96 mg, 32%) $R_f$ 0.45 (EtOAc); mp 212-216 °C; $\nu_{max}$ (neat) 3112 (C-H), 2994 (C-H), 2954 (C-H), 2867 (C-H), 2838 (C-H), 1723 (ester C=O), 1643 (ketone C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 2.57 (s, 3 H, C(15)$H_3$), 3.33 - 3.48 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 3.79 - 3.94 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 4.00 (s, 3 H, C(25)*H*), 6.70 (dd, *J*=8.0, 3.0 Hz, 1 H, C(3)*H*), 7.55 (dd, *J*=5.0, 1.5 Hz, 1 H, C(21)*H*), 7.88 (s, 1 H, C(17)*H*), 8.12 (d, *J*=3.0 Hz, 1 H, C(5)*H*), 8.15 (s, 1 H, C(23)*H*), 8.74 (d, *J*=5.0 Hz, 1 H, C(20)*H*), 9.76 (d, *J*=8.0 Hz, 1 H, C(2)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 26.8 (s, 1 C, *C*(15)), 47.5 (s, 2 C, *C*(8)+*C*(12)), 52.6 (s, 1 C, *C*(25)), 66.5 (s, 2 C, *C*(9)+*C*(11)), 99.6 (s, 1 C, *C*(5)), 106.0 (s, 1 C, *C*(3)), 111.4 (s, 1 C, *C*(16)), 118.0 (s, 1 C, *C*(21)), 118.9 (s, 1 C, *C*(23)), 121.4 (s, 1 C, *C*(13)), 123.6 (s, 1 C, *C*(17)), 129.7 (s, 1 C, *C*(2)), 137.4 (s, 1 C, *C*(22)), 139.5 (s, 1 C, *C*(6)), 148.9 (s, 1 C, *C*(4)), 149.8 (s, 1 C, *C*(20)), 155.9 (s, 1 C, *C*(18)), 166.1 (s, 1 C, *C*(24)), 185.3 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 380 [MH$^+$]; HRMS (ESI$^+$) found 402.1417, calculated for $C_{21}H_{21}N_3NaO_4^+$ 402.1424; HPLC (System D) $t_r$ 15.0 min (95%).

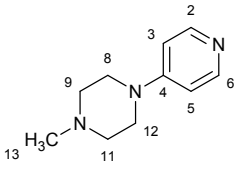
## 2-[3-Acetyl-7-(morpholin-4-yl)indolizin-1-yl]pyridine-4-carboxylic acid (23)



1.0 M aq. LiOH solution (0.26 mL, 0.26 mmol) was added to a stirred suspension of compound **22** (50 mg, 0.13 mmol) in THF (1.0 mL) and MeOH (1 mL). The mixture was heated at 50 °C for 2 h then allowed to cool. The reaction mixture was neutralised by addition of 1 M aq. HCl. The resultant precipitate was collected by filtration, washed with a small volume of MeOH, then dried under vacuum to yield the product as a brick-red

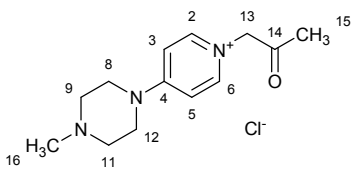
solid (37 mg, 78%); $R_f$ 0.10 ($CH_2Cl_2$:MeOH, 80:20); mp 212-217 °C; $\nu_{max}$ (neat) 3118 (C-H), 2947 (C-H), 2875 (C-H), 1713 (acid C=O) 1646 (ketone C=O); $^1$H NMR (500 MHz, DMSO-$d_6$) $\delta$ ppm 2.53 (s, 3 H, C(15)$H_3$) 3.25 - 3.47 (m, 4 H+$H_2O$, C(8)$H_2$+C(12)$H_2$) 3.71 - 3.87 (m, 4 H, C(9)$H_2$+C(11)$H_2$) 7.09 (dd, *J*=8.0, 3.0 Hz, 1 H, C(3)*H*) 7.51 (dd, *J*=5.0, 1.5 Hz, 1 H, C(21)*H*) 8.18 (d, *J*=3.0 Hz, 1 H, C(5)*H*) 8.29 (s, 1 H, C(23)*H*) 8.40 (s, 1 H, C(17)*H*) 8.73 (d, *J*=5.0 Hz, 1 H, C(20)*H*) 9.64 (d, *J*=8.0 Hz, 1 H, C(2)*H*); $^{13}$C NMR (126 MHz, DMSO-$d_6$) $\delta$ ppm 26.6 (s, 1 C, *C*(15)), 46.8 (s, 2 C, *C*(8)+*C*(12)), 65.8 (s, 2 C, *C*(9)+*C*(11)), 98.8 (s, 1 C, *C*(5)), 106.4 (s, 1 C, *C*(3)), 110.6 (s, 1 C, *C*(16)), 118.1 (s, 1 C, *C*(21)), 118.7 (s, 1 C, *C*(23)), 120.8 (s, 1 C, *C*(13)), 124.4 (s, 1 C, *C*(17)), 128.7 (s, 1 C, *C*(2)), 138.9 (s, 1 C, *C*(22)), 139.7 (s, 1 C, *C*(6)), 148.6 (s, 1 C, *C*(4)), 149.7 (s, 1 C, *C*(20)), 155.5 (s, 1 C, *C*(18)), 166.8 (s, 1 C, *C*(24)), 185.0 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 366 [MH$^+$], 364 [(M-H)$^-$]; HRMS found 366.1443, calculated for $C_{20}H_{20}N_3O_4^+$ 366.1448; HPLC (System D) $t_r$ 11.4 min (98%).

### 1-Methyl-4-(pyridin-4-yl)piperazine




Iodomethane (6.15 g, 43.3 mmol) was added drop-wise to a stirred suspension of 1-(piperidine-4-yl)piperazine (7.00 g, 42.9 mmol) and $K_2CO_3$ (11.9 g, 85.8 mmol) in DMF (200 mL). The resultant mixture was stirred at room temperature for 4 h then quenched by addition of *conc*. $NH_4OH$ (0.5 mL). The mixture was concentrated *in vacuo* then partitioned between EtOAc (200 mL) and water (200 mL). The phases were separated then the aqueous phase was extracted with EtOAc (5 × 200 mL). The combined organic phases were washed with brine (200 mL) then dried over $MgSO_4$ and evaporated. The material thus obtained was re-dissolved in MeOH then loaded onto a pre-wetted SCX cartridge (70 g, Biotage). The cartridge was eluted with MeOH then the basic components were recovered through elution with 7 M $NH_3$ in MeOH. The ammonia eluent was evaporated to yield the product as a pale-yellow oil (4.24 g, 56%); $R_f$ 0.25 ($CH_2Cl_2$:MeOH:$NH_4OH$, 90:10:1); $\nu_{max}$ (neat) 3030 (C-H), 2939 (C-H), 2843 (C-H), 2797 (C-H); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 2.27 (s, 3 H, C(13)$H_3$), 2.40 - 2.50 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 3.25 - 3.31 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 6.54 - 6.64 (m, 2 H, C(3)*H*+C(5)*H*), 8.16 - 8.25 (m, 2 H, C(2)*H*+C(6)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 45.8 (s, 2 C, *C*(9)+*C*(11)), 46.1 (s, 1 C, *C*(13)), 54.4 (s, 2 C, *C*(8)+*C*(12)), 108.3 (s, 2 C, *C*(3)+*C*(5)), 150.1 (s, 2 C, *C*(2)+*C*(6)), 154.8 (s, 1 C, *C*(4)); LRMS *m/z* (ESI$^+$) 200 [(M+Na)$^+$], 178 [MH$^+$]; HRMS (ESI$^+$) found 178.1339, calculated for $C_{10}H_{16}N_3^+$ 178.1339; HPLC (System E) $t_r$ 0.4 min (>99%).

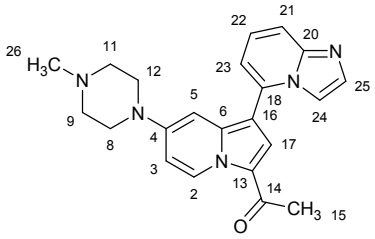
### 4-(4-Methylpiperazin-1-yl)-1-(2-oxopropyl)pyridinium chloride (6)




Chloroacetone (90 µL, 1.1 mmol) was added drop-wise to a stirred solution of 1-methyl-4-(pyridin-4-yl)piperazine (200 mg, 1.1 mmol) in THF (6 mL). The resultant mixture was left to stir at room temperature for 18 h. The resultant suspension was filtered then the solid was washed with THF and dried under vacuum to yield the product as a pale-yellow solid (104 mg, 34%); mp 238-242 °C; $\nu_{max}$ (neat) 3036 (C-H), 2988 (C-H),

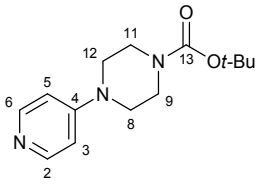
2939 (C-H), 2839 (C-H), 2787 (C-H), 2750 (C-H), 2707 (C-H), 1730 (C=O); $^1$H NMR (400 MHz, DMSO-$d_6$) δ ppm 2.06 - 2.26 (m, 6 H, C(15)$H_3$+C(16)$H_3$), 2.36 - 2.47 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 3.51 - 3.77 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 5.36 (s, 2 H, C(13)$H_2$), 7.30 (d, *J*=8.0 Hz, 2 H, C(3)*H*+C(5)*H*), 8.14 (d, *J*=8.0 Hz, 2 H, C(2)*H*+C(6)*H*); $^{13}$C NMR (101 MHz, DMSO-$d_6$) δ ppm 26.9 (s, 1 C, *C*(15)), 45.3 (s, 1 C, *C*(16)), 45.9 (s, 2 C, *C*(8)+*C*(12)), 53.9 (s, 2 C, *C*(9)+*C*(11)), 64.5 (s, 1 C, *C*(13)), 107.6 (s, 2 C, *C*(3)+*C*(5)), 143.6 (s, 2 C, *C*(2)+*C*(6)), 155.4 (s, 1 C, *C*(4)), 201.0 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 234 [M$^+$]; HRMS (ESI$^+$) found 234.1610, calculated for $C_{13}H_{20}N_3O^+$ 234.1601; HPLC (System E) $t_r$ 0.6 min (>99%).

### 1-[1-(Imidazo[1,2-*a*]pyridin-5-yl)-7-(4-methylpiperazin-1-yl)indolizin-3-yl]ethanone (31)



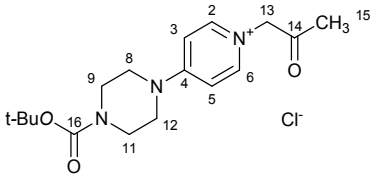

$K_2CO_3$ (138 mg, 1.00 mmol) was added to a solution of compound **6** (135 mg, 0.50 mmol) in DMF (5 mL). The resultant suspension was stirred at room temperature for 15 minutes then 5-ethynylimidazo[1,2*a*]pyridine (78 mg, 0.55 mmol) was added. The mixture was then heated at 90 °C for 4 h, allowed to cool, then partitioned between water (10 mL) and $CHCl_3$ (10 mL). The organic phase was collected by passing it through a hydrophobic frit then concentrated by nitrogen blow-down. The crude material was purified by flash column chromatography on a silica column (24 g). The column was eluted with a gradient of $CH_2Cl_2$:-MeOH:$NH_4OH$ which was increased linearly from 99:1:0.1 to 95:5:0.5 over 10 CVs. The desired fractions were combined and evaporated to a red gum (39 mg). The material thus obtained was purified further by flash column chromatography on a C-18 column (13 g). The column was eluted with a gradient of $H_2O$:MeCN (+0.1% $CF_3CO_2H$) which was increased linearly from 95:5 to 5:95 over 20 CVs. The desired fractions were combined and evaporated then partitioned between $CHCl_3$ (5 mL) and saturated aq. $NaHCO_3$ (5 mL). The organic phase was collected by passing through a hydrophobic frit then evaporated to a red gum (19 mg). The material thus obtained was re-dissolved in the minimum of $CHCl_3$ then loaded onto a preparative TLC plate (Analtech, 20×20 cm, 2000 μm). Eluted with $CH_2Cl_2$:MeOH:$NH_4OH$ (95:5:0.5). The desired band was scratched from the TLC plate with a spatula then suspended in $CH_2Cl_2$:MeOH:$NH_4OH$ (90:10:1). The mixture was filtered then the filtrate was evaporated to yield the product as a tan-brown residue (5 mg, 2.7%); $R_f$ 0.45 ($CH_2Cl_2$:MeOH:$NH_4OH$, 90:10:1); $\nu_{max}$ (neat) 3096 (C-H), 2925 (C-H), 2875 (C-H), 2841 (C-H), 2803 (C-H), 2771 (C-H), 1645 (C=O); $^1$H NMR (500 MHz, $CDCl_3$) δ ppm 2.37 (s, 3 H, C(26)$H_3$), 2.55 (s, 3 H, C(15)$H_3$), 2.56 - 2.60 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 3.27 - 3.32 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 6.45 (d, *J*=2.5 Hz, 1 H, C(5)*H*), 6.75 (dd, *J*=8.0, 2.5 Hz, 1 H, C(3)*H*), 6.89 (d, *J*=7.0 Hz, 1 H, C(23)*H*), 7.28 - 7.34 (m, 1 H, C(22)*H*), 7.61 - 7.73 (m, 4 H, C(17)*H*+C(21)*H*+C(24)*H*+C(25)*H*), 9.79 (d, *J*=8.0 Hz, 1 H, C(2)*H*); $^{13}$C NMR (126 MHz, $CDCl_3$) δ ppm 26.7 (s, 1 C, *C*(15)), 45.9 (s, 1 C, *C*(26)), 47.1 (s, 2 C, *C*(8)+*C*(12)), 54.3 (s, 2 C, *C*(9)+*C*(11)), 96.6 (s, 1 C, *C*(5)), 105.8 (s, 1 C, *C*(16)), 106.5 (s, 1 C, *C*(3)), 111.6 (s, 1 C, *C*(24)), 113.0 (s, 1 C, *C*(23)), 115.6 (s, 1 C, *C*(21)), 121.5 (s, 1 C, *C*(13)), 124.5 (s, 1 C, *C*(17)), 124.7 (s, 1 C, *C*(22)), 130.1 (s, 1 C, *C*(2)), 132.9 (s, 1 C, *C*(18)), 133.0 (s, 1 C, *C*(25)), 138.5 (s, 1 C, *C*(6)), 146.2 (s, 1 C, *C*(20)), 148.0 (s, 1 C, *C*(4)), 185.4 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 374 [MH$^+$]; HRMS (ESI$^+$) found 374.1980, calculated for $C_{22}H_{24}N_5O^+$ 374.1975; HPLC (System D) $t_r$ 7.7 min (91%).

### *tert*-Butyl 4-(pyridin-4-yl)piperazine-1-carboxylate



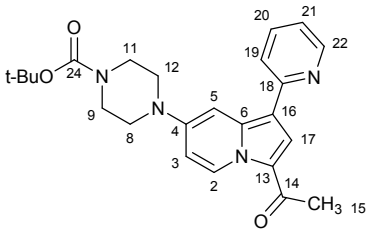

Di-*tert*-butyl dicarbonate (2.18 g, 10.0 mmol) was added to a solution of 1-(pyridine-4-yl)piperazine (1.63 g, 10.0 mmol) in THF (50 mL). The resultant solution was left to stir at room temperature for 2 h then concentrated *in vacuo*. The residue was partitioned between EtOAc (50 mL) and water (50 mL). The phases were separated then the organic phase was washed with brine (50 mL) then dried over $MgSO_4$ and evaporated to yield the product as a white solid (2.34 g, 89%); $R_f$ 0.30 ($CH_2Cl_2$:MeOH:$NH_4OH$, 90:10:1); mp 99-104 °C; $\nu_{max}$ (neat) 2978 (C-H), 2862 (C-H), 1686 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 1.48 (s, 9 H, 9×Boc-*H*), 3.27 - 3.42 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 3.47 - 3.59 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 6.61 - 6.71 (m, 2 H, C(3)*H*+C(5)*H*), 8.28 (d, *J*=6.5 Hz, 2 H, C(2)*H*+C(6)*H*)); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 28.3 (s, 3 C, 3×Boc-*C*) 42.9 (s, 2 C, *C*(9)+*C*(11)), 45.8 (s, 2 C, *C*(8)+*C*(12)), 80.2 (s, 1 C, Boc-*C*) 108.5 (s, 2 C, *C*(3)+*C*(5)), 150.3 (s, 2 C, *C*(2)+*C*(6)), 154.5 (s, 1 C, *C*(4/13)), 154.7 (s, 1 C, *C*(4/13)); LRMS *m/z* (ESI$^+$) 264 [MH$^+$]; HRMS (ESI$^+$) found 264.1695, calculated for $C_{22}H_{24}N_5O^+$ 264.1707; HPLC (System D) $t_r$ 9.1 min (98%).

### 4-[4-(*tert*-Butoxycarbonyl)piperazin-1-yl]-1-(2-oxopropyl)pyridinium chloride (7)



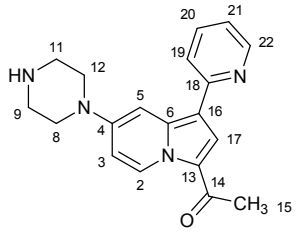

Chloroacetone (0.233 mL, 2.85 mmol) was added to a stirred solution of *tert*-butyl 4-(pyridin-4-yl)piperazine-1-carboxylate (750 mg, 2.85 mmol) in THF (20 mL). The resultant mixture was left to stir for 18 h at room temperature then diluted with $Et_2O$ (20 mL). The solid was filtered then dried under vacuum to yield the product as a pale-yellow solid (662 mg, 65%); mp 229-231 °C; $\nu_{max}$ (neat) 3034 (C-H), 2982 (C-H), 2921 (C-H), 2866 (C-H), 1735 (ketone C=O), 1692 (carbamate C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 1.44 (s, 9 H, 9×Boc-*H*), 2.34 (s, 3 H, C(15)$H_3$), 3.56 - 3.66 (m, 4 H, C(8/9)$H_2$+C(11/12)$H_2$), 3.66 - 3.80 (m, 4 H, C(8/9)$H_2$+C(11/12)$H_2$), 5.87 - 6.01 (m, 2 H, C(13)$H_2$), 7.07 - 7.19 (m, 2 H, C(3)*H*+C(5)*H*), 8.42 - 8.57 (m, 2 H, C(2)*H*+C(6)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 27.6 (s, 1 C, *C*(15)), 28.2 (s, 3 C, 3×Boc-*C*) 46.1 (s, 4 C, *C*(8)+*C*(9)+*C*(11)+*C*(12)), 65.3 (s, 1 C, *C*(13)), 80.7 (s, 1 C, Boc-*C*) 107.9 (s, 2 C, *C*(3)+*C*(5)), 144.3 (s, 2 C, *C*(2)+*C*(6)), 154.2 (s, 1 C, *C*(4/16)), 155.9 (s, 1 C, *C*(4/16)), 200.7 (s, 1 C, *C*(21)); LRMS *m/z* (ESI$^+$) 320 [M$^+$]; HRMS (ESI$^+$) found 320.1954, calculated for $C_{17}H_{26}N_3O_3^+$ 320.1969; HPLC (System D) $t_r$ 9.2 min (91%).

### *tert*-Butyl 4-[3-acetyl-1-(pyridin-2-yl)indolizin-7-yl]piperazine-1-carboxylate



A suspension of $K_2CO_3$ (40 mg, 0.29 mmol) was added to a solution of compound **7** (93 mg, 0.26 mmol) in DMF (2 mL). The resultant suspension was stirred at room temperature for 10 minutes then 2-ethynylpyridine (29 µL, 0.29 mmol) was added. The mixture was then heated at 90 °C for 2 h then allowed to cool. The mixture was then partitioned between water (5 mL) and $CH_2Cl_2$ (5 mL). The organic phase was collected by passing it through a hydrophobic frit then evaporated. The crude material was purified by flash column chromatography on a silica column (4 g). The column was eluted with a gradient of EtOAc:*c*-hexane + 0.1% $NEt_3$ which was increased linearly from 10:90 to 50:50 over 20 CVs. The desired fractions were combined and evaporated to yield the product as a yellow resin (23 mg, 21%); $R_f$ 0.25 (EtOAc:*c*-hexane, 60:40 + 1% $NEt_3$); $\nu_{max}$ (neat) 3107 (C-H), 3049 (C-H), 2979 (C-H), 2923 (C-H), 2822 (C-H), 1696 (carbamate C=O), 1639 (ketone C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 1.49 (s, 9 H, 9×Boc-*H*), 2.55 (s, 3 H, C(15)$H_3$), 3.30 - 3.47 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 3.50 - 3.68 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 6.69 (dd, *J*=8.0, 2.5 Hz, 1 H, C(3)*H*), 7.07 (ddd, *J*=7.5, 5.0, 1.0 Hz, 1 H, C(21)*H*), 7.57 - 7.65 (m, 1 H, C(19)*H*), 7.65 - 7.73 (m, 1 H, C(20)*H*), 7.81 (s, 1 H, C(17)*H*), 8.04 (d, *J*=2.5 Hz, 1 H, C(5)*H*), 8.57 - 8.67 (m, 1 H, C(22)*H*), 9.76 (d, *J*=8.0 Hz, 1 H, C(2)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 26.7 (s, 1 C, *C*(15)), 28.4 (s, 3 C, 3×Boc-*C*) 42.7 (s, 2 C, *C*(9)+*C*(11)), 47.5 (s, 2 C, *C*(8)+*C*(12)), 80.1 (s, 1 C, Boc-*C*) 100.0 (s, 1 C, *C*(5)), 106.6 (s, 1 C, *C*(3)), 112.2 (s, 1 C, *C*(16)), 119.6 (s, 1 C, *C*(19/21)), 119.9 (s, 1 C, *C*(19/21)), 121.3 (s, 1 C, *C*(13)), 123.5 (s, 1 C, *C*(17)), 129.7 (s, 1 C, *C*(2)), 136.3 (s, 1 C, *C*(20)), 139.2 (s, 1 C, *C*(6)), 148.5 (s, 1 C, *C*(4)), 149.0 (s, 1 C, *C*(22)), 154.6 (s, 1 C, *C*(22/24)), 154.7 (s, 1 C, *C*(22/24)), 185.1 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 443 ([M+Na]$^+$), 421 [MH$^+$]; HRMS (ESI$^+$) found 421.2238, calculated for $C_{24}H_{29}N_4O_3^+$ 421.2234; HPLC (System D) $t_r$ 11.7 min (89%).

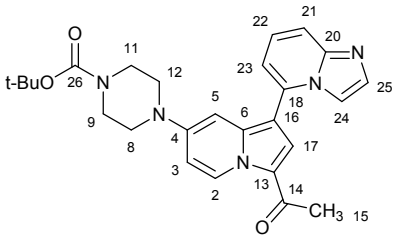
### 1-[7-(Piperazin-1-yl)-1-(pyridin-2-yl)indolizin-3-yl]ethanone dihydrochloride (29)



4 M HCl in dioxane (1 mL, 4 mmol) was added to a stirred suspension of *tert*-butyl 4-[3-acetyl-1-(pyridin-2-yl)indolizin-7-yl]piperazine-1-carboxylate (35 mg, 0.083 mmol) in dioxane (1 mL). The suspension was stirred at room temperature for 16 h then evaporated to yield the product as a yellow solid (33 mg, quant.); mp 210 °C (dec) $\nu_{max}$ (neat) 3356 (N-H), 2805 (C-H), 1644 (C=O); $^1$H NMR (500 MHz, $CD_3OD$) $\delta$ ppm 2.62 (s, 3 H, C(15)$H_3$), 3.35 - 3.55 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 3.77 - 3.90 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 7.17 (d, *J*=8.0 Hz, 1 H, C(3)*H*), 7.39 (s, 1 H, C(5)*H*), 7.72 – 7.87 (m, 1 H, C(21)*H*), 8.28 (s, 1 H, C(17)*H*), 8.39 (d, *J*=8.0 Hz, 1 H, C(19)*H*), 8.48 - 8.60 (m, 1 H, C(20)*H*), 8.66 (d, *J*=5.5 Hz, 1 H, C(22)*H*), 9.78 (d, *J*=8.0 Hz, 1 H, C(2)*H*); $^{13}$C NMR (126 MHz,

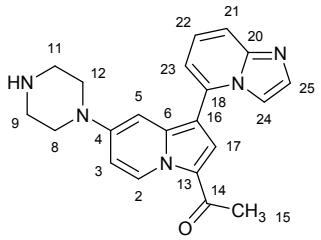
$CD_3OD$) δ ppm 26.9 (s, 1 C, *C*(15)), 44.4 (s, 2 C, *C*(9)+*C*(11)), 45.4 (s, 2 C, *C*(8)+*C*(12)), 97.8 (s, 1 C, *C*(5)), 104.6 (s, 1 C, *C*(16)), 108.4 (s, 1 C, *C*(3)), 123.0 (s, 1 C, *C*(21)), 124.8 (s, 1 C, *C*(13)), 126.4 (s, 1 C, *C*(19)), 126.9 (s, 1 C, *C*(17)), 131.9 (s, 1 C, *C*(2)), 140.8 (s, 1 C, *C*(6)), 141.9 (s, 1 C, *C*(22)), 147.3 (s, 1 C, *C*(20)), 149.5 (s, 1 C, *C*(4)), 150.9 (s, 1 C, *C*(18)), 188.7 (s, 1 C, *C*(14)); LRMS *m/z* ($ESI^+$) 321 [$MH^+$]; HRMS ($ESI^+$) found 321.1700, calculated for $C_{19}H_{21}N_4O^+$ 321.1710; HPLC (System D) $t_r$ 7.5 min (99%).

### *tert*-Butyl 4-[3-acetyl-1-(imidazo[1,2-*a*]pyridin-5-yl)indolizin-7-yl]piperazine-1-carboxylate



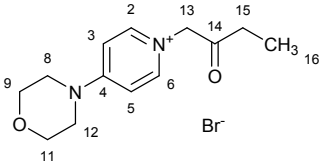

A suspension of $K_2CO_3$ (66 mg, 0.48 mmol) was added to a solution of compound **7** (155 mg, 0.44 mmol) in DMF (5 mL). The resultant suspension was stirred at room temperature for 5 minutes then 5-ethynylimidazo[1,2*a*]pyridine (68 mg, 0.48 mmol) was added. The mixture was then heated at 90 °C for 2 h, allowed to cool, then concentrated *in vacuo*. The residue was then partitioned between water (5 mL) and EtOAc (5 mL). The aqueous phase was extracted with more EtOAc (5 mL) then the combined organic phases were washed water (5 mL) and brine (5 mL) then dried over $MgSO_4$ and evaporated. The crude material was purified by flash column chromatography on a silica column (12 g). The column was eluted with a gradient of $CH_2Cl_2$:MeOH:$NH_4OH$ which was increased linearly from 99:1:0.1 to 90:10:1 over 12 CVs. The desired fractions were combined and evaporated to yield a yellow gum (132 mg). The material thus obtained was purified further by flash column chromatography on a C-18 column (13 g). The column was eluted with a gradient of $H_2O$:MeCN (+0.1% $CF_3CO_2H$) which was increased linearly from 95:5 to 5:95 over 20 CVs. The desired fractions were combined and evaporated then partitioned between $CH_2Cl_2$ (5 mL) and saturated aq. $NaHCO_3$ (5 mL). The organic phase was collected by passing through a hydrophobic frit then evaporated to yield the product as a yellow gum (74 mg, 20%); $R_f$ 0.50 ($CH_2Cl_2$:MeOH:$NH_4OH$, 90:10:1); $\nu_{max}$ (neat) 3105 (C-H), 2975 (C-H), 2922 (C-H), 2853 (C-H), 1689 (carbamate C=O), 1644 (ketone C=O); $^1$H NMR (400 MHz, $CDCl_3$) δ ppm 1.46 (s, 9 H, 9×Boc-*H*), 2.55 (s, 3 H, C(15)$H_3$), 3.09 - 3.33 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 3.47 - 3.68 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 6.44 (d, *J*=2.5 Hz, 1 H, C(5)*H*), 6.75 (dd, *J*=8.0, 2.5 Hz, 1 H, C(3)*H*), 6.93 (d, *J*=7.0 Hz, 1 H, C(23)*H*), 7.36 (dd, *J*=9.0, 7.0 Hz, 1 H, C(22)*H*), 7.50 - 7.81 (m, 4 H, C(17)*H*+C(21)*H*+C(24)*H*+C(25)*H*), 9.79 (d, *J*=8.0 Hz, 1 H, C(2)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) δ ppm 26.7 (s, 1 C, *C*(15)), 28.3 (s, 3 C, 3×Boc-*C*) 43.0 (s, 2 C, *C*(9)+*C*(11)), 47.3 (s, 2 C, *C*(8)+*C*(12)), 80.3 (s, 1 C, Boc-*C*) 96.7 (s, 1 C, *C*(5)), 105.4 (s, 1 C, *C*(16)), 106.8 (s, 1 C, *C*(3)), 111.8 (s, 1 C, *C*(24)), 113.5 (s, 1 C, *C*(23)), 115.2 (s, 1 C, *C*(21)), 121.6 (s, 1 C, *C*(13)), 124.4 (s, 1 C, *C*(17)), 125.6 (s, 1 C, *C*(22)), 130.1 (s, 1 C, *C*(2)), 131.8 (s, 1 C, *C*(18)), 133.0 (s, 1 C, *C*(25)), 138.4 (s, 1 C, *C*(6)), 146.2 (s, 1 C, *C*(20)), 148.1 (s, 1 C, *C*(4)), 154.4 (s, 1 C, *C*(26)), 185.6 (s, 1 C, *C*(14)); LRMS *m/z* ($ESI^+$) 460 [$MH^+$]; HRMS ($ESI^+$) found 460.2358, calculated for $C_{26}H_{30}N_5O_3^+$ 460.2343; HPLC (System E) $t_r$ 4.4 min (92%).

**1-[1-(Imidazo[1,2-*a*]pyridin-5-yl)-7-(piperazin-1-yl)indolizin-3-yl]ethanone dihydrochloride (30)**



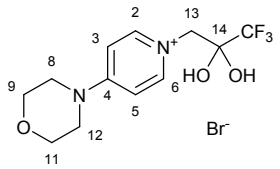
4 M HCl in dioxane (2 mL, 8 mmol) was added to a stirred suspension of *tert*-butyl 4-[3-acetyl-1-(imidazo[1,2-*a*]pyridin-5-yl)indolizin-7-yl]piperazine-1-carboxylate (70 mg, 0.15 mmol) in dioxane (2 mL) and MeOH (0.5 mL). The suspension was stirred at room temperature for 1 h then evaporated to yield the product as a dark green solid (66 mg, quant.); mp 320 °C (dec); $\nu_{max}$ (neat) 3473 (N-H), 3383 (N-H), 2914 (C-H), 2833 (C-H), 2757 (C-H), 2635 (C-H), 1648 (C=O); $^1$H NMR (500 MHz, $CD_3OD$) $\delta$ ppm 2.49 (s, 3 H, C(15)$H_3$), 3.24 - 3.31 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 3.50 - 3.58 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 6.77 (d, *J*=2.5 Hz, 1 H, C(5)*H*), 7.01 (dd, *J*=8.0, 2.5 Hz, 1 H, C(3)*H*), 7.53 (dd, *J*=7.5, 0.5 Hz, 1 H, C(23)*H*), 7.81 (d, *J*=9.0 Hz, 1 H, C(21)*H*), 7.95 - 7.99 (m, 2 H, C(22)*H*+C(25)*H*), 7.99 (s, 1 H, C(17)*H*), 8.03 (d, *J*=1.5 Hz, 1 H, C(24)*H*), 9.68 (d, *J*=8.0 Hz, 1 H, C(2)*H*); $^{13}$C NMR (126 MHz, $CD_3OD$) $\delta$ ppm 26.9 (s, 1 C, *C*(15)), 44.3 (s, 2 C, *C*(9)+*C*(11)), 45.7 (s, 2 C, *C*(8)+*C*(12)), 98.3 (s, 1 C, *C*(5)), 104.6 (s, 1 C, *C*(16)), 108.6 (s, 1 C, *C*(3)), 110.7 (s, 1 C, *C*(21)), 115.5 (s, 1 C, *C*(24)), 119.2 (s, 1 C, *C*(23)), 123.7 (s, 1 C, *C*(25)), 123.7 (s, 1 C, *C*(13)), 127.2 (s, 1 C, *C*(17)), 131.5 (s, 1 C, *C*(2)), 135.3 (s, 1 C, *C*(22)), 137.2 (s, 1 C, *C*(18/20)), 140.7 (s, 1 C, *C*(6)), 142.7 (s, 1 C, *C*(18/20)), 149.8 (s, 1 C, *C*(4)), 188.4 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 360 [MH$^+$]; HRMS (ESI$^+$) found 360.1814, calculated for $C_{21}H_{22}N_5O^+$ 360.1819; HPLC (System E) $t_r$ 2.7 min (95%).

**4-(Morpholin-4-yl)-1-(2-oxobutyl)pyridinium bromide (8)**



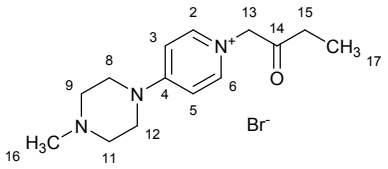
1-Bromo-2-butanone (311 μL, 3.05 mmol) was added to a stirred solution of 4-morpholinopyridine (500 mg, 3.05 mmol) in THF (15 mL). The resultant mixture was left to stir for 1 h at room temperature then the solid precipitate was filtered and dried under vacuum to yield the product as a white solid (858 mg, 89%); mp 320-325°C; $\nu_{max}$ (neat) 3036 (C-H), 2996 (C-H), 2902 (C-H), 2857 (C-H), 1721 (C=O); $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 1.00 (t, *J*=7.0 Hz, 3 H, C(16)$H_3$), 2.59 (q, *J*=7.0 Hz, 2 H, C(15)$H_2$), 3.59 - 3.80 (m, 8 H+solvent, C(8)$H_2$+C(9)$H_2$+C(11)$H_2$+ C(12)$H_2$), 5.29 (s, 2 H, C(13)$H_2$), 7.24 (d, *J*=8.0 Hz, 2 H, C(3)*H*+C(5)*H*), 8.08 (d, *J*=8.0 Hz, 2 H, C(2)*H*+C(6)*H*); $^{13}$C NMR (101 MHz, DMSO-$d_6$) $\delta$ ppm 7.3 (s, 1 C, *C*(16)), 32.6 (s, 1 C, *C*(15)), 46.3 (s, 2 C, *C*(8)+*C*(12)), 64.2 (s, 1 C, *C*(13)), 65.8 (s, 2 C, *C*(9)+*C*(11)), 107.8 (s, 2 C, *C*(3)+*C*(5)), 143.9 (s, 2 C, *C*(2)+*C*(6)), 156.0 (s, 1 C, *C*(4)), 204.0 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 235 [M$^+$]; HRMS (ESI$^+$) found 235.1443, calculated for $C_{13}H_{19}N_2O_2^+$ 235.1441; HPLC (System E) $t_r$ 2.7 min (90%).

## 4-(Morpholin-4-yl)-1-(3,3,3-trifluoro-2,2-dihydroxypropyl)pyridinium bromide (9)



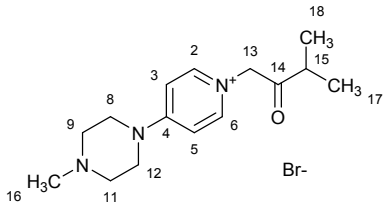
3-Bromo-1,1,1-trifluoroacetone (632 µL, 6.09 mmol) was added to a stirred solution of 4-morpholinopyridine (1.00 g, 6.09 mmol) in THF (15 mL). The resultant mixture was left to stir for 1 h at room temperature then filtered to remove the precipitate. The filtrate was left to stand for 64 h, resulting in further precipitation. The supernatant was decanted off then the solid was suspended in acetone, filtered, and dried under vacuum to yield the product as a cream solid (337 mg, 15%); mp 235-239 °C; $\nu_{max}$ (neat) 3213 (O-H), 3055 (C-H); $^{1}$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 3.63 - 3.86 (m, 8 H, C(8)$H_2$+C(9)$H_2$+C(11)$H_2$+C(12)$H_2$), 4.46 (s, 2 H, C(13)$H_2$), 7.29 (d, *J*=8.0 Hz, 2 H, C(3)*H*+C(5)*H*), 7.72 (s, 2 H, 2×O*H*), 8.21 (d, *J*=8.0 Hz, 2 H, C(2)*H*+C(6)*H*); $^{13}$C NMR (101 MHz, DMSO-$d_6$) $\delta$ ppm 46.1 (s, 2 C, *C*(8)+*C*(12)), 58.8 (s, 1 C, *C*(13)), 65.5 (s, 2 C, *C*(9)+*C*(11)), 91.2 (q, *J*=31.0 Hz, 1 C, *C*(14)), 107.2 (s, 2 C, *C*(3)+*C*(5)), 123.2 (q, *J*=285.0 Hz, 1 C, *C*$F_3$) 144.4 (s, 2 C, *C*(2)+*C*(6)), 155.9 (s, 1 C, *C*(4)); $^{19}$F NMR (377 MHz, DMSO-$d_6$) $\delta$ ppm -82.40 (s, 3 F); LRMS *m/z* (ESI$^+$) 307 [(M-$H_2O$+MeOH)$^+$], 293 [M$^+$]; HRMS (ketone) (ESI$^+$) found 275.1001, calculated for $C_{12}H_{14}F_3N_2O_2^+$ 275.1002; HRMS (MeOH adduct) (ESI$^+$) found 307.1263, calculated for $C_{15}H_{18}F_3N_2O_3^+$ 307.1264; HPLC (System E) $t_r$ 2.5 min (98%).

## 4-(4-Methylpiperazin-1-yl)-1-(2-oxobutyl)pyridinium bromide (10)



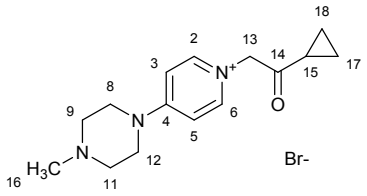
1-Bromo-2-butanone (115 µL, 1.13 mmol) was added drop-wise to a stirred solution of 1-methyl-4-(pyridin-4-yl)piperazine (200 mg, 1.13 mmol) in THF (6 mL). The resultant mixture was left to stir at room temperature for 18 h. The resultant suspension was filtered then the solid was washed with THF and dried under vacuum to yield the product as a pale-yellow solid (268 mg, 72%); mp 255-259 °C; $\nu_{max}$ (neat) 3038 (C-H), 2977 (C-H), 2940 (C-H), 2909 (C-H), 2954 (C-H), 2790 (C-H), 2745 (C-H), 1725 (C=O); $^{1}$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 1.02 (t, *J*=7.5 Hz, 3 H, C(17)$H_3$), 2.24 (s, 3 H, C(16)$H_3$), 2.41 - 2.50 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 2.59 (q, *J*=7.5 Hz, 2 H, C(15)$H_2$), 3.61 - 3.78 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 5.32 (s, 2 H, C(13)$H_2$), 7.31 (d, *J*=8.0 Hz, 2 H, C(3)*H*+C(5)*H*), 8.12 (d, *J*=8.0 Hz, 2 H, C(2)*H*+C(6)*H*); $^{13}$C NMR (101 MHz, DMSO-$d_6$) $\delta$ ppm 7.0 (s, 1 C, *C*(17)), 32.3 (s, 1 C, *C*(15)), 45.3 (s, 1 C, *C*(16)), 45.9 (s, 2 C, *C*(8)+*C*(12)), 53.9 (s, 2 C, *C*(9)+*C*(11)), 63.8 (s, 1 C, *C*(13)), 107.6 (s, 2 C, *C*(3)+*C*(5)), 143.6 (s, 2 C, *C*(2)+*C*(6)), 155.4 (s, 1 C, *C*(4)), 203.6 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 248 [M$^+$]; HRMS (ESI$^+$) found 248.1756, calculated for $C_{14}H_{22}N_3O^+$ 248.1757; HPLC (System E) $t_r$ 0.8 min (>99%).

## 1-(3-Methyl-2-oxobutyl)-4-(4-methylpiperazin-1-yl)pyridinium bromide (11)



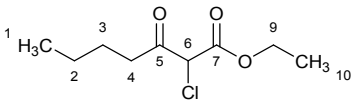

1-Bromo-3-methyl-2-butanone (489 mg, 2.96 mmol) was added drop-wise to a stirred solution of 1-methyl-4-(pyridin-4-yl)piperazine (500 mg, 2.82 mmol) in THF (15 mL). The resultant mixture was left to stir at room temperature for 4 h. The resultant suspension was filtered then the solid was washed with acetone and dried under vacuum to yield the product as a pale-yellow solid (672 mg, 70%); mp 198-201°C; $\nu_{max}$ (neat) 3034 (C-H), 2936 (C-H), 2797 (C-H), 1721 (C=O); $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 1.13 (d, *J*=7.0 Hz, 6 H, C(17)$H_3$+C(18)$H_3$), 2.26 (s, 3 H, C(16)$H_3$), 2.43 - 2.52 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 2.80 (spt, *J*=7.0 Hz, 1 H, C(15)*H*), 3.69 - 3.79 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 5.48 (s, 2 H, C(13)$H_2$), 7.32 (d, *J*=8.0 Hz, 2 H, C(3)*H*+C(5)*H*), 8.17 (d, *J*=8.0 Hz, 2 H, C(2)*H*+C(6)*H*); $^{13}$C NMR (101 MHz, DMSO-$d_6$) $\delta$ ppm 17.6 (s, 2 C, *C*(17)+*C*(18)), 37.7 (s, 1 C, *C*(15)), 45.1 (s, 1 C, *C*(16)), 45.8 (s, 2 C, *C*(8)+*C*(12)), 53.8 (s, 2 C, *C*(9)+*C*(11)), 62.7 (s, 1 C, *C*(13)), 107.7 (s, 2 C, *C*(3)+*C*(5)), 143.6 (s, 2 C, *C*(2)+*C*(6)), 155.4 (s, 1 C, *C*(4)), 206.6 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 262 [M$^+$]; HRMS (ESI$^+$) found 262.1918, calculated for $C_{15}H_{24}N_3O^+$ 262.1914; HPLC (System D) $t_r$ 2.2 min (83%).

## 1-(2-Cyclopropyl-2-oxoethyl)-4-(4-methylpiperazin-1-yl)pyridinium bromide (12)



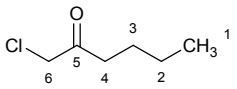

2-Bromo-1-cyclopropylethanone (483 mg, 2.96 mmol) was added drop-wise to a stirred solution of 1-methyl-4-(pyridin-4-yl)piperazine (500 mg, 2.82 mmol) in THF (15 mL). The resultant mixture was left to stir at room temperature for 4 h. The resultant suspension was filtered then the solid was washed with acetone and dried under vacuum to yield the product as an off-white solid (427 mg, 44%); mp 233-237 °C; $\nu_{max}$ (neat) 3034 (C-H), 2936 (C-H), 2903 (C-H), 2793 (C-H), 1707 (C=O); $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 0.95 - 1.12 (m, 4 H, C(17)$H_2$+C(18)$H_2$), 2.14 - 2.23 (m, 1 H, C(15)*H*), 2.26 (s, 3 H, C(16)$H_3$), 2.44 - 2.50 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 3.69 - 3.76 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 5.51 (s, 2 H, C(13)$H_2$), 7.30 (d, *J*=8.0 Hz, 2 H, C(3)*H*+C(5)*H*), 8.16 (d, *J*=8.0 Hz, 2 H, C(2)*H*+C(6)*H*); $^{13}$C NMR (101 MHz, DMSO-$d_6$) $\delta$ ppm 11.2 (s, 2 C, *C*(17)+*C*(18)), 18.3 (s, 1 C, *C*(15)), 45.1 (s, 1 C, *C*(16)), 45.8 (s, 2 C, *C*(8)+*C*(12)), 53.8 (s, 2 C, *C*(9)+*C*(11)), 64.2 (s, 1 C, *C*(13)), 107.6 (s, 2 C, *C*(3)+*C*(5)), 143.6 (s, 2 C, *C*(2)+*C*(6)), 155.4 (s, 1 C, *C*(4)), 202.9 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 260 [M$^+$]; HRMS (ESI$^+$) found 260.1761, calculated for $C_{15}H_{22}N_3O^+$ 260.1757; HPLC (System D) $t_r$ 2.3 min (73%).

**Ethyl 2-chloro-3-oxoheptanoate**[2,3]



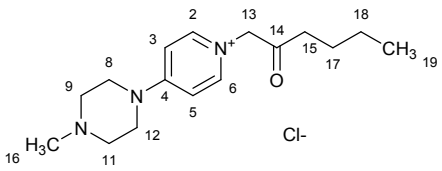
$SO_2Cl_2$ (426 µL, 5.25 mmol) was added drop-wise to a stirred solution of ethyl 3-oxoheptanoate (800 µL, 5.00 mmol) in $CH_2Cl_2$ (4 mL). The resultant solution was left to stir at room temperature for 1 h then added to water (5 mL). The phases were separated then the organic phase was washed with water (5 mL) and brine (5 mL) then dried by passing it through a hydrophobic frit. The dried solution was evaporated to yield the product as a colourless oil (1.01 g, 98%); $R_f$ 0.50 (EtOAc:*c*-hexane, 20:80); $\nu_{max}$ (neat) 2962 (C-H), 2937 (C-H), 2875 (C-H), 1729 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 0.92 (t, *J*=7.5 Hz, 3 H, C(1)$H_3$), 1.26 - 1.45 (m, 5 H, C(2)$H_2$+C(10)$H_3$), 1.62 (quin, *J*=7.5 Hz, 2 H, C(3)$H_2$), 2.71 (q, *J*=7.5 Hz, 2 H, C(4)$H_2$), 4.29 (q, *J*=7.0 Hz, 2 H, C(9)$H_2$), 4.78 (s, 1 H, C(6)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 13.7 (s, 1 C, *C*(1)), 13.9 (s, 1 C, *C*(10)), 22.0 (s, 1 C, *C*(2)), 25.5 (s, 1 C, *C*(3)), 38.6 (s, 1 C, *C*(4)), 60.9 (s, 1 C, *C*(6)), 63.1 (s, 1 C, *C*(9)), 165.1 (s, 1 C, *C*(7)), 199.1 (s, 1 C, *C*(5)); LRMS *m/z* (ESI$^+$) 229 [(M+Na)$^+$] 224 [(M+$H_2O$)$^+$]; HRMS (ESI$^+$) found 229.0610, calculated for $C_9H_{15}ClNaO_3^+$ 229.0602.

**1-Chlorohexan-2-one**[4]



A mixture of ethyl 2-chloro-3-oxoheptanoate in $H_2O$ (2.4 g) and $H_2SO_4$(2.4 g) was heated under reflux for 24 h. The reaction mixture was added onto $H_2O$ (15 mL) then extracted with $CH_2Cl_2$ (2×15 mL). The combined organic phases were dried over $MgSO_4$ and evaporated to a brown oil (415 mg, 64%); $\nu_{max}$ (neat) 2961 (C-H), 2934 (C-H), 2874 (C-H), 1739 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 0.85 (t, *J*=7.5 Hz, 3 H, C(1)$H_3$), 1.27 (sxt, *J*=7.5 Hz, 2 H, C(2)$H_2$), 1.54 (quin, *J*=7.5 Hz, 1 H, C(3)$H_2$), 2.52 (t, *J*=7.5 Hz, 2 H, C(4)$H_2$), 4.01 (s, 2 H, C(6)$H_2$); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 14.2 (s, 1 C, *C*(1)), 22.6 (s, 1 C, *C*(2)), 26.1 (s, 1 C, *C*(3)), 39.9 (s, 1 C, *C*(4)), 48.7 (s, 1 C, *C*(6)), 203.2 (s, 1 C, *C*(5)).

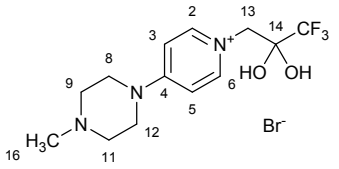
**4-(4-Methylpiperazin-1-yl)-1-(2-oxohexyl)pyridinium chloride (13)**



1-Chlorohexan-2-one (228 mg, 1.69 mmol) was added drop-wise to a stirred solution of 1-methyl-4-(pyridin-4-yl)piperazine (300 mg, 1.69 mmol) in THF (10 mL). The resultant mixture was left to stir at room temperature for 18 h. The resultant suspension was filtered then the filtrate was left to stand for 24 h, resulting in further precipitation. The precipitate was filtered, to yield the product as a beige solid (427 mg, 44%); mp 210-214 °C; $\nu_{max}$ (neat) 3036 (C-H), 2948 (C-H), 2869 (C-H), 2810 (C-H), 2778 (C-H), 1727 (C=O); $^1$H NMR (500 MHz, DMSO-$d_6$) $\delta$ ppm 0.89 (t, *J*=7.5 Hz, 3 H, C(19)$H_3$), 1.31 (sxt, *J*=7.5 Hz, 2 H, C(18)$H_2$), 1.52 (quin, *J*=7.5 Hz, 2 H, C(17)$H_2$), 2.24 (s, 3 H, C(16)$H_3$), 2.41 - 2.50 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 2.57 (t, *J*=7.5 Hz, 2 H,

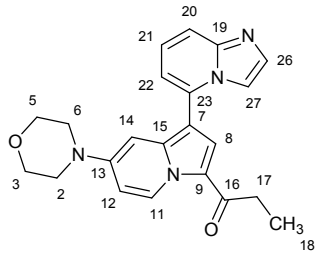
C(15)$H_2$), 3.66 - 3.76 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 5.30 (s, 2 H, C(13)$H_2$), 7.29 (d, *J*=8.0 Hz, 2 H, C(3)*H*+C(5)*H*), 8.11 (d, *J*=8.0 Hz, 2 H, C(2)*H*+C(6)*H*); $^{13}$C NMR (126 MHz, DMSO-$d_6$) δ ppm 13.8 (s, 1 C, *C*(19)), 21.7 (s, 1 C, *C*(18)), 24.7 (s, 1 C, *C*(17)), 38.6 (s, 1 C, *C*(15)), 45.3 (s, 1 C, *C*(16)), 45.9 (s, 2 C, *C*(8)+*C*(12)), 53.9 (s, 2 C, *C*(9)+*C*(11)), 64.0 (s, 1 C, *C*(13)), 107.6 (s, 2 C, *C*(3)+*C*(5)), 143.6 (s, 2 C, *C*(2)+*C*(6)), 155.4 (s, 1 C, *C*(4)), 203.2 (s, 1 C, *C*(14)); LRMS *m/z* (ESI$^+$) 276 [M$^+$]; HRMS (ESI$^+$) found 276.2063, calculated for $C_{16}H_{26}N_3O^+$ 276.2070; HPLC (System D) $t_r$ 2.4 min (76%).

### 4-(4-Methylpiperazin-1-yl)-1-(3,3,3-trifluoro-2,2-dihydroxypropyl)pyridinium bromide (14)




3-Bromo-1,1,1-trifluoroacetone (293 μL, 2.82 mmol) was added to a stirred solution of 1-methyl-4-(pyridin-4-yl)piperazine (500 mg, 2.82 mmol) in acetone (15 mL). The resultant mixture was left to stir for 16 h at room temperature then filtered. The solid was washed with acetone then dried under vacuum to yield the product as a beige solid (261 mg, 25%); mp 188-192 °C; $\nu_{max}$ (neat) 2986 (C-H), 2963 (C-H), 2868 (C-H); $^1$H NMR (400 MHz, DMSO-$d_6$) δ ppm 2.31 (s, 3 H, C(16)$H_3$), 2.53 - 2.59 (m, 4 H, C(9)$H_2$+C(11)$H_2$), 3.72 - 3.79 (m, 4 H, C(8)$H_2$+C(12)$H_2$), 4.45 (s, 2 H, C(13)$H_2$), 7.30 (d, *J*=8.0 Hz, 2 H, C(3)*H*+C(5)*H*), 7.72 (br. s., 2 H, 2×O*H*), 8.19 (d, *J*=8.0 Hz, 2 H, C(2)*H*+C(6)*H*); $^{13}$C NMR (101 MHz, DMSO-$d_6$) δ ppm 44.9 (s, 1 C, *C*(16)), 45.6 (s, 2 C, *C*(8)+*C*(12)), 53.6 (s, 2 C, *C*(9)+*C*(11)), 58.7 (s, 1 C, *C*(13)), 91.2 (q, *J*=31.5 Hz, 1 C, *C*(14)), 107.3 (s, 2 C, *C*(3)+*C*(5)), 123.1 (q, *J*=285.0 Hz, 1 C, *C*F$_3$) 144.4 (s, 2 C, *C*(2)+*C*(6)), 155.7 (s, 1 C, *C*(14)); $^{19}$F NMR (377 MHz, DMSO-$d_6$) δ ppm -82.42 (s, 3 F); LRMS *m/z* (ESI$^+$) 320 [(M-$H_2$O+MeOH)$^+$], 306 [M$^+$]; HRMS (ESI$^+$) found 306.1423, calculated for $C_{13}H_{19}F_3N_3O_2^+$ 306.1424; HPLC (System D) $t_r$ 1.5 min (>99%).

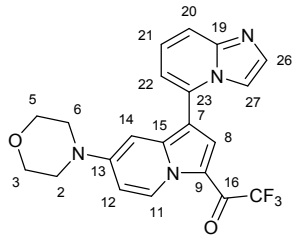
### 1-[1-(Imidazo[1,2-*a*]pyridin-5-yl)-7-(morpholin-4-yl)indolizin-3-yl]propan-1-one (32)




$K_2CO_3$ (111 mg, 0.80 mmol) was added to a solution of compound **8** (126 mg, 0.40 mmol) in DMF (5 mL). The resultant suspension was stirred at room temperature for 15 minutes then 5-ethynylimidazo[1,2*a*]pyridine (63 mg, 0.44 mmol) was added. The mixture was then heated at 90 °C for 3 h then allowed to cool. The resultant mixture was partitioned between water (5 mL) and EtOAc (5 mL). The phases were separated then the aqueous phase was extracted with more EtOAc (2×5 mL). The combined organic phases were washed with water (2×5mL) and brine (5 mL) then dried over $MgSO_4$ and evaporated. The crude material was pre-adsorbed onto silica then purified by flash column chromatography on a silica column (12 g). The column was eluted with a gradient of $CH_2Cl_2$:MeOH:$NH_4OH$ which was increased linearly from 99:1:0.1 to 95:5:0.5 over 12 CVs. The desired fractions were combined and evaporated to a beige solid (56 mg). The material thus obtained was purified further by flash column chromatography on a C-18 column (13 g). The column

was eluted with a gradient of $H_2O$:MeCN (+0.1% $CF_3CO_2H$) which was increased linearly from 95:5 to 5:95 over 20 CVs. The desired fractions were combined and evaporated then partitioned between $CHCl_3$ (5 mL) and saturated aq. $NaHCO_3$ (5 mL). The organic phase was collected by passing through a hydrophobic frit then evaporated to yield the product as a yellow gum (5 mg, 3.3%); $R_f$ 0.55 ($CH_2Cl_2$:MeOH:$NH_4OH$, 90:10:1); $\nu_{max}$ (neat) 3071 (C-H), 2973 (C-H), 2943 (C-H), 2868 (C-H), 1644 (C=O); $^1$H NMR (700 MHz, $CDCl_3$) $\delta$ ppm 1.30 (t, *J*=7.5 Hz, 3 H, C(18)$H_3$), 2.91 (q, *J*=7.5 Hz, 2 H, C(17)$H_2$), 3.12 - 3.23 (m, 4 H, C(2)$H_2$+C(6)$H_2$), 3.76 - 3.86 (m, 4 H, C(3)$H_2$+C(5)$H_2$), 6.46 (d, *J*=2.5 Hz, 1 H, C(14)*H*), 6.75 (dd, *J*=8.0, 2.5 Hz, 1 H, C(12)*H*), 6.90 (d, *J*=7.0 Hz, 1 H, C(22)*H*), 7.31 (dd, *J*=9.0, 7.0 Hz, 1 H, C(21)*H*), 7.59 (s, 1 H, C(27)*H*), 7.65 - 7.69 (m, 2 H, C(20)*H*+C(26)*H*), 7.72 (s, 1 H, C(8)*H*), 9.84 (d, *J*=8.0 Hz, 1 H, C(11)*H*); $^{13}$C NMR (176 MHz, $CDCl_3$) $\delta$ ppm 9.7 (s, 1 C, *C*(18)), 32.0 (s, 1 C, *C*(17)), 47.5 (s, 2 C, *C*(2)+*C*(6)), 66.3 (s, 2 C, *C*(3)+*C*(5)), 96.5 (s, 1 C, *C*(14)), 105.7 (s, 1 C, *C*(7)), 106.2 (s, 1 C, *C*(12)), 111.6 (s, 1 C, *C*(27)), 113.1 (s, 1 C, *C*(22)), 115.6 (s, 1 C, *C*(20) 121.1 (s, 1 C, *C*(9)), 123.6 (s, 1 C, *C*(8)), 124.8 (s, 1 C, *C*(21)), 130.1 (s, 1 C, *C*(11)), 132.8 (s, 1 C, *C*(26)), 132.9 (s, 1 C, *C*(23)), 138.3 (s, 1 C, *C*(15)), 146.1 (s, 1 C, *C*(19)), 148.2 (s, 1 C, *C*(13)), 189.4 (s, 1 C, *C*(16)); LRMS *m/z* (ESI$^+$) 375 [MH$^+$]; HRMS (ESI$^+$) found 375.1806, calculated for $C_{22}H_{23}N_4O_2^+$ 375.1816; HPLC (System E) $t_r$ 4.3 min (97%).

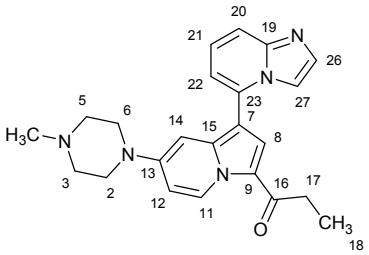
### 2,2,2-Trifluoro-1-[1-(imidazo[1,2-*a*]pyridin-5-yl)-7-(morpholin-4-yl)indolizin-3-yl]ethanone (33)




$K_2CO_3$ (111 mg, 0.80 mmol) was added to a solution of compound **9** (149 mg, 0.40 mmol) in DMF (5 mL). The resultant suspension was stirred at room temperature for 15 minutes then 5-ethynylimidazo[1,2*a*]pyridine (63 mg, 0.44 mmol) was added. The mixture was then heated at 90 °C for 3 h then allowed to cool. The resultant mixture was partitioned between water (5 mL) and EtOAc (5 mL). The phases were separated then the aqueous phase was extracted with more EtOAc (2×5 mL). The combined organic phases were washed with water (2×5mL) and brine (5 mL) then dried over $MgSO_4$ and evaporated. The crude material was purified by flash column chromatography on a silica column (12 g). The column was eluted with a gradient of $CH_2Cl_2$:MeOH:$NH_4OH$ which was increased linearly from 99:1:0.1 to 95:5:0.5 over 12 CVs. The desired fractions were combined and evaporated to yield the product as a yellow residue (24 mg, 14%); $R_f$ 0.50 ($CH_2Cl_2$:MeOH:$NH_4OH$, 90:10:1); $\nu_{max}$ (neat) 2960 (C-H), 2854 (C-H), 1649 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 3.23 - 3.36 (m, 4 H, C(2)$H_2$+C(6)$H_2$), 3.79 - 3.87 (m, 4 H, C(3)$H_2$+C(5)$H_2$), 6.51 (d, *J*=2.5 Hz, 1 H, C(14)*H*), 6.87 (dd, *J*=8.0, 2.5 Hz, 1 H, C(12)*H*), 6.91 (dd, *J*=7.0, 1.0 Hz, 1 H, C(22)*H*), 7.30 (dd, *J*=9.0, 7.0 Hz, 1 H, C(21)*H*), 7.49 - 7.55 (m, 1 H, C(27)*H*), 7.66 - 7.72 (m, 2 H, C(20)*H*+C(26)*H*), 7.86 (q, *J*=2.0 Hz, 1 H, C(8)*H*), 9.79 (d, *J*=8.0 Hz, 1 H, C(11)*H*); $^{13}$C NMR (176 MHz, $CDCl_3$) $\delta$ ppm 46.8 (s, 2 C, *C*(2)+*C*(6)), 66.1 (s, 2 C, *C*(3)+*C*(5)), 96.6 (s, 1 C, *C*(14)), 106.4 (s, 1 C, *C*(12)), 109.7 (s, 1 C, *C*(7)), 111.4 (s, 1 C, *C*(27)), 113.6 (s, 1 C, *C*(22)), 117.8 (q, *J*=289.5 Hz, 1 C, *C*$F_3$) 116.5 (s, 1 C, *C*(9)), 116.6 (s, 1 C, *C*(20)), 124.4 (s, 1 C, *C*(21)), 126.9 (q, *J*=4.0 Hz, 1 C, *C*(8)), 131.0 (s, 1 C, *C*(11)), 131.4 (s, 1 C, *C*(23)), 133.4 (s, 1 C, *C*(26)), 141.3 (s, 1 C, *C*(15)), 146.1 (s, 1 C, *C*(19)), 150.1 (s, 1 C, *C*(13)), 166.0 (q, *J*=35.0 Hz, 1 C, *C*(16)); $^{19}$F NMR (377 MHz, $CDCl_3$) $\delta$ ppm -70.22 (s, 3 F); LRMS *m/z* (ESI$^+$) 415

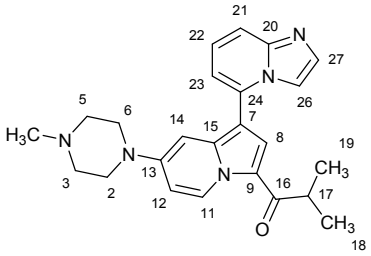
[MH$^+$]; HRMS (ESI$^+$) found 415.1379, calculated for $C_{21}H_{18}F_3N_4O_2$ 415.1376; HPLC (System D) $t_r$ 10.7 min (99%).

### 1-[1-(Imidazo[1,2-*a*]pyridin-5-yl)-7-(4-methylpiperazin-1-yl)indolizin-3-yl]propan-1-one (34)



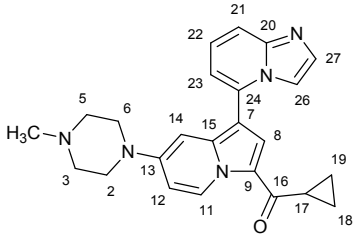
$K_2CO_3$ (111 mg, 0.80 mmol) was added to a solution of compound **10** (131 mg, 0.40 mmol) in DMF (5 mL). The resultant suspension was stirred at room temperature for 15 minutes then 5-ethynylimidazo[1,2*a*]pyridine (63 mg, 0.44 mmol) was added. The mixture was then heated at 90 °C for 3 h then allowed to cool and stirred at room temperature for 16 h. The resultant mixture was partitioned between water (5 mL) and EtOAc (5 mL). The phases were separated then the aqueous phase was extracted with more EtOAc (2×5 mL). The combined organic phases were washed with water (2×5mL) and brine (5 mL) then dried over $MgSO_4$ and evaporated. The crude material was purified by flash column chromatography on a silica column (12 g). The column was eluted with a gradient of $CH_2Cl_2$:MeOH:$NH_4OH$ which was increased linearly from 99:1:0.1 to 90:10:1 over 20 CVs. The desired fractions were combined and evaporated to a brown/orange gum (28 mg). The material thus obtained was purified further by flash column chromatography on a silica column (12 g). The column was eluted with a gradient of $CH_2Cl_2$:MeOH:$NH_4OH$ which was increased linearly from 96:4:0.4 to 93:7:0.7 over 12 CVs. The desired fractions were combined and evaporated then the material thus obtained was purified once more by column chromatography silica column (4 g). The column was eluted isocratically with EtOAc:MeOH:$NEt_3$ (90:10:1). The desired fractions were combined and evaporated to yield the product as a pale-orange solid (15 mg, 10%); $R_f$ 0.25 ($CH_2Cl_2$:-MeOH:$NH_4OH$, 90:10:1); $\nu_{max}$ (neat) 2977 (C-H), 2941 (C-H), 2807 (C-H), 1645 (C=O);$^1$H NMR (700 MHz, $CDCl_3$) $\delta$ ppm 1.29 (t, *J*=7.5 Hz, 3 H, C(18)$H_3$), 2.34 (s, 3 H, NC$H_3$), 2.50 - 2.58 (m, 4 H, C(3)$H_2$+C(5)$H_2$), 2.89 (q, *J*=7.5 Hz, 2 H, C(17)$H_2$), 3.22 - 3.30 (m, 4 H, C(2)$H_2$+C(6)$H_2$), 6.44 (d, *J*=2.5 Hz, 1 H, C(14)*H*), 6.75 (dd, *J*=8.0, 2.5 Hz, 1 H, C(12)*H*), 6.87 (d, *J*=7.0 Hz, 1 H, C(22)*H*), 7.28 (dd, *J*=9.0, 7.0 Hz, 1 H+solvent, C(21)*H*), 7.58 (s, 1 H, C(27)*H*), 7.63 (d, *J*=9.0 Hz, 1 H, C(20)*H*), 7.65 (s, 1 H, C(26)*H*), 7.70 (s, 1 H, C(8)*H*), 9.81 (d, *J*=8.0 Hz, 1 H, C(11)*H*); $^{13}$C NMR (176 MHz, $CDCl_3$) $\delta$ ppm 9.7 (s, 1 C, *C*(18)), 31.9 (s, 1 C, *C*(17)), 46.0 (s, 1 C, NC$H_3$) 47.2 (s, 2 C, *C*(2)+*C*(6)), 54.4 (s, 2 C, *C*(3)+*C*(5)), 96.6 (s, 1 C, *C*(14)), 105.7 (s, 1 C, *C*(7)), 106.4 (s, 1 C, *C*(12)), 111.6 (s, 1 C, *C*(27)), 112.8 (s, 1 C, *C*(22)), 115.6 (s, 1 C, *C*(20)), 121.0 (s, 1 C, *C*(9)), 123.6 (s, 1 C, *C*(8)), 124.5 (s, 1 C, *C*(21)), 130.0 (s, 1 C, *C*(11)), 132.9 (s, 1 C, *C*(23)), 133.1 (s, 1 C, *C*(26)), 138.4 (s, 1 C, *C*(15)), 146.3 (s, 1 C, *C*(19)), 148.0 (s, 1 C, *C*(13)), 189.2 (s, 1 C, *C*(16)); LRMS *m/z* (ESI$^+$) 388 [MH$^+$]; HRMS (ESI$^+$) found 388.2135, calculated for $C_{23}H_{26}N_5O$ 388.2132; HPLC (System D) $t_r$ 8.2 min (99%).

**1-[1-(Imidazo[1,2-*a*]pyridin-5-yl)-7-(4-methylpiperazin-1-yl)indolizin-3-yl]-2-methylpropan-1-one (35)**



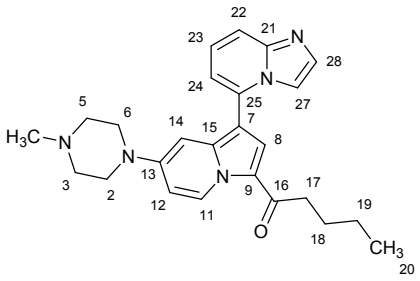
$K_2CO_3$ (138 mg, 1.00 mmol) was added to a solution of compound **11** (171 mg, 0.50 mmol) in DMF (5 mL). The resultant suspension was stirred at room temperature for 10 minutes then 5-ethynylimidazo[1,2*a*]pyridine (78 mg, 0.55 mmol) was added. The mixture was then heated at 90 °C for 4 h then allowed to cool. The resultant mixture was partitioned between water (10 mL) and $CHCl_3$ (10 mL). The phases were separated then the organic phase was evaporated under a stream of nitrogen. The crude material was purified by flash column chromatography on a silica column (24 g). The column was eluted with a gradient of $CH_2Cl_2$:MeOH:$NH_4OH$ which was increased linearly from 99:1:0.1 to 95:5:0.5 over 10 CVs. The desired fractions were combined and evaporated to an red gum (54 mg). The material thus obtained was purified further by flash column chromatography on a C-18 column (13 g). The column was eluted with a gradient of $H_2O$:MeCN (+0.1% $CF_3CO_2H$) which was increased linearly from 95:5 to 5:95 over 20 CVs. The desired fractions were combined and evaporated then partitioned between $CHCl_3$ (5 mL) and saturated aq. $NaHCO_3$ (5 mL). The organic phase was collected by passing through a hydrophobic frit then evaporated to yield the product as a yellow gum (41 mg, 20%); $R_f$ 0.45 ($CH_2Cl_2$:MeOH:$NH_4OH$, 90:10:1); $\nu_{max}$ (neat) 2967 (C-H), 2936 (C-H), 2844 (C-H), 2799 (C-H), 1644 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 1.27 (d, *J*=7.0 Hz, 6 H, C(18)$H_3$+C(19)$H_3$), 2.34 (s, 3 H, NC$H_3$), 2.50 - 2.59 (m, 4 H, C(3)$H_2$+C(5)$H_2$), 3.22 - 3.30 (m, 4 H, C(2)$H_2$+C(6)$H_2$), 3.40 (spt, *J*=7.0 Hz, 1 H, C(17)*H*), 6.45 (d, *J*=2.5 Hz, 1 H, C(14)*H*), 6.75 (dd, *J*=8.0, 2.5 Hz, 1 H, C(12)*H*), 6.88 (dd, *J*=7.0, 1.0 Hz, 1 H, C(23)*H*), 7.28 (dd, *J*=9.0, 7.0 Hz, 1 H+solvent, C(22)*H*), 7.58 - 7.60 (m, 1 H, C(26)*H*), 7.61 - 7.67 (m, 2 H, C(25)*H*+C(27)*H*), 7.73 (s, 1 H, C(8)*H*), 9.84 (d, *J*=8.0 Hz, 1 H, C(11)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 20.1 (s, 2 C, *C*(18)+*C*(19)), 36.2 (s, 1 C, *C*(17)), 46.0 (s, 1 C, N*C*$H_3$) 47.2 (s, 2 C, *C*(2)+*C*(6)), 54.4 (s, 2 C, *C*(3)+*C*(5)), 96.6 (s, 1 C, *C*(14)), 105.7 (s, 1 C, *C*(7)), 106.4 (s, 1 C, *C*(12)), 111.6 (s, 1 C, *C*(26)), 112.8 (s, 1 C, *C*(23)), 115.6 (s, 1 C, *C*(21)), 120.3 (s, 1 C, *C*(9)), 123.5 (s, 1 C, *C*(8)), 124.5 (s, 1 C, *C*(22)), 130.2 (s, 1 C, *C*(11)), 132.9 (s, 1 C, *C*(24)), 133.2 (s, 1 C, *C*(27)), 138.7 (s, 1 C, *C*(15)), 146.3 (s, 1 C, *C*(20)), 148.1 (s, 1 C, *C*(13)), 192.9 (s, 1 C, *C*(16)); LRMS *m/z* (ESI$^+$) 402 [MH$^+$]; HRMS (ESI$^+$) found 402.2297, calculated for $C_{24}H_{28}N_5O$ 402.2288; HPLC (System D) $t_r$ 8.5 min (95%).

**Cyclopropyl[1-(imidazo[1,2-*a*]pyridin-5-yl)-7-(4-methylpiperazin-1-yl)indolizin-3-yl]methanone (36)**



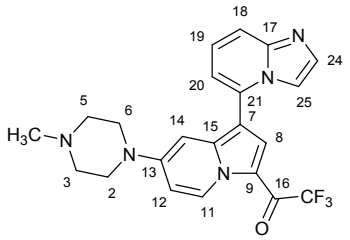
$K_2CO_3$ (138 mg, 1.00 mmol) was added to a solution of compound **12** (170 mg, 0.50 mmol) in DMF (5 mL). The resultant suspension was stirred at room temperature for 10 minutes then 5-ethynylimidazo[1,2*a*]pyridine (78 mg, 0.55 mmol) was added. The mixture was then heated at 90 °C for 4 h then allowed to cool. The resultant mixture was partitioned between water (10 mL) and $CHCl_3$ (10 mL). The phases were separated then the organic phase was evaporated under a stream of nitrogen. The crude material was purified by flash column chromatography on a silica column (24 g). The column was eluted with a gradient of $CH_2Cl_2$:MeOH:$NH_4OH$ which was increased linearly from 99:1:0.1 to 95:5:0.5 over 10 CVs. The desired fractions were combined and evaporated to a red gum (52 mg). The material thus obtained was purified further by flash column chromatography on a C-18 column (13 g). The column was eluted with a gradient of $H_2O$:MeCN (+0.1% $CF_3CO_2H$) which was increased linearly from 95:5 to 5:95 over 20 CVs. The desired fractions were combined and evaporated then partitioned between $CHCl_3$ (5 mL) and saturated aq. $NaHCO_3$ (5 mL). The organic phase was collected by passing through a hydrophobic frit then evaporated to yield the product as a red/brown gum (28 mg, 14%); $R_f$ 0.45 ($CH_2Cl_2$:MeOH:$NH_4OH$, 90:10:1); $\nu_{max}$ (neat) 3088 (C-H), 3004 (C-H), 2941 (C-H), 2844 (C-H), 1642 (C=O); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ ppm 0.89 - 0.99 (m, 2 H, C(18)$H_AH_B$+C(19)$H_AH_B$), 1.18 - 1.26 (m, 2 H, C(18)$H_AH_B$+C(19)$H_AH_B$), 2.35 (s, 3 H, NC$H_3$), 2.45 - 2.58 (m, 5 H, C(3)$H_2$+C(5)$H_2$+C(17)*H*), 3.24 - 3.32 (m, 4 H, C(2)$H_2$+C(6)$H_2$), 6.46 (d, *J*=2.5 Hz, 1 H, C(14)*H*), 6.73 (dd, *J*=8.0, 2.5 Hz, 1 H, C(12)*H*), 6.90 (dd, *J*=7.0, 1.0 Hz, 1 H, C(23)*H*), 7.30 (dd, *J*=9.0, 7.0 Hz, 1 H+solvent, C(22)*H*), 7.60 - 7.68 (m, 3 H, C(21)*H*+C(26)*H*+C(27)*H*), 7.85 (s, 1 H, C(8)*H*), 9.80 (d, *J*=8.0 Hz, 1 H, C(11)*H*); $^{13}$C NMR (101 MHz, $CDCl_3$) $\delta$ ppm 9.7 (s, 2 C, *C*(18)+*C*(19)), 17.5 (s, 1 C, *C*(17)), 46.0 (s, 1 C, N*C*$H_3$) 47.2 (s, 2 C, *C*(2)+*C*(6)), 54.4 (s, 2 C, *C*(3)+*C*(5)), 96.5 (s, 1 C, *C*(14)), 105.9 (s, 1 C, *C*(7)), 106.4 (s, 1 C, *C*(12)), 111.6 (s, 1 C, *C*(21)), 112.9 (s, 1 C, *C*(23)), 115.6 (s, 1 C, *C*(21)), 122.0 (s, 1 C, *C*(9)), 123.7 (s, 1 C, *C*(8)), 124.6 (s, 1 C, *C*(22)), 130.0 (s, 1 C, *C*(11)), 133.0 (s, 1 C, *C*(24)), 133.1 (s, 1 C, *C*(27)), 138.4 (s, 1 C, *C*(15)), 146.3 (s, 1 C, *C*(20)), 147.9 (s, 1 C, *C*(13)), 187.4 (s, 1 C, *C*(16)); LRMS *m/z* (ESI$^+$) 400 [MH$^+$]; HRMS (ESI$^+$) found 400.2129, calculated for $C_{24}H_{26}N_5O$ 400.2132; HPLC (System D) $t_r$ 8.4 min (95%).

## 1-[1-(Imidazo[1,2-*a*]pyridin-5-yl)-7-(4-methylpiperazin-1-yl)indolizin-3-yl]propan-1-one (37)



$K_2CO_3$ (53 mg, 0.38 mmol) was added to a solution of compound **13** (60 mg, 0.19 mmol) in DMF (2 mL). The resultant suspension was stirred at room temperature for 10 minutes then 5-ethynylimidazo[1,2*a*]pyridine (30 mg, 0.21 mmol) was added. The mixture was then heated at 90 °C for 4 h then allowed to cool. The resultant mixture was partitioned between water (5 mL) and $CHCl_3$ (5 mL). The organic phase was collected by passing it through a hydrophobic frit, then evaporated using a stream of nitrogen. The crude material was purified by flash column chromatography on a silica column (12 g). The column was eluted with a gradient of $CH_2Cl_2$:MeOH:$NH_4OH$ which was increased linearly from 99:1:0.1 to 95:5:0.5 over 10 CVs, then isocratic at 95:5:0.5 for 5 CVs. The desired fractions were combined and evaporated to an orange gum (15 mg). The material thus obtained was purified further by flash column chromatography on a C-18 column (13 g). The column was eluted with a gradient of $H_2O$:MeCN (+0.1% $CF_3CO_2H$) which was increased linearly from 95:5 to 5:95 over 20 CVs. The desired fractions were combined and evaporated then partitioned between $CHCl_3$ (5 mL) and saturated aq. $NaHCO_3$ (5 mL). The organic phase was collected by passing through a hydrophobic frit then evaporated to yield the product as a brown gum (6 mg, 7.6%); $R_f$ 0.45 ($CH_2Cl_2$:MeOH:$NH_4OH$, 90:10:1); $\nu_{max}$ (neat) 2955 (C-H), 2932 (C-H), 2844 (C-H), 2802 (C-H), 1644 (C=O); $^1$H NMR (500 MHz, $CDCl_3$) $\delta$ ppm 0.97 (t, *J*=7.5 Hz, 3 H, C(20)$H_3$), 1.44 (sxt, *J*=7.5 Hz, 2 H, C(19)$H_2$), 1.79 (quin, *J*=7.5 Hz, 2 H, C(18)$H_2$), 2.37 (s, 3 H, NC$H_3$), 2.54 - 2.64 (m, 4 H, C(3)$H_2$+C(5)$H_2$), 2.86 (t, *J*=7.5 Hz, 2 H, C(17)$H_2$), 3.23 - 3.33 (m, 4 H, C(2)$H_2$+C(6)$H_2$), 6.46 (d, *J*=2.5 Hz, 1 H, C(14)*H*), 6.75 (dd, *J*=8.0, 2.5 Hz, 1 H, C(12)*H*), 6.89 (dd, *J*=7.0, 1.0 Hz, 1 H, C(24)*H*), 7.30 (dd, *J*=9.0, 7.0 Hz, 1 H, C(23)*H*), 7.59 (s, 1 H, C(27)*H*), 7.63 - 7.68 (m, 2 H, C(22)*H*+C(28)*H*), 7.70 (s, 1 H, C(8)*H*), 9.83 (d, *J*=8.0 Hz, 1 H, C(11)*H*); $^{13}$C NMR (126 MHz, $CDCl_3$) $\delta$ ppm 14.0 (s, 1 C, *C*(20)), 22.7 (s, 1 C, *C*(19)), 28.2 (s, 1 C, *C*(18)), 38.8 (s, 1 C, *C*(17)), 45.9 (s, 1 C, N*C*$H_3$) 47.2 (s, 2 C, *C*(2)+*C*(6)), 54.3 (s, 2 C, *C*(3)+*C*(5)), 96.6 (s, 1 C, *C*(14)), 105.7 (s, 1 C, *C*(7)), 106.5 (s, 1 C, *C*(12)), 111.6 (s, 1 C, *C*(27)), 113.0 (s, 1 C, *C*(24)), 115.6 (s, 1 C, *C*(22)), 121.4 (s, 1 C, *C*(9)), 123.9 (s, 1 C, *C*(8)), 124.7 (s, 1 C, *C*(23)), 130.1 (s, 1 C, *C*(11)), 133.0 (s, 1 C, *C*(25)), 133.0 (s, 1 C, *C*(28)), 138.5 (s, 1 C, *C*(15)), 146.3 (s, 1 C, *C*(21)), 148.0 (s, 1 C, *C*(13)), 188.9 (s, 1 C, *C*(16)); LRMS *m/z* (ESI$^+$) 416 [MH$^+$]; HRMS (ESI$^+$) found 416.2448, calculated for $C_{25}H_{30}N_5O$ 416.2445; HPLC (System D) $t_r$ 9.0 min (92%).

**2,2,2-Trifluoro-1-[1-(imidazo[1,2-*a*]pyridin-5-yl)-7-(4-methylpiperazin-1-yl)indolizin-3-yl]ethanone (38)**



$K_2CO_3$ (138 mg, 1.00 mmol) was added to a solution of compound **14** (193 mg, 0.50 mmol) in DMF (5 mL). The resultant suspension was stirred at room temperature for 10 minutes then 5-ethynylimidazo[1,2*a*]pyridine (78 mg, 0.55 mmol) was added. The mixture was then heated at 90 °C for 4 h then allowed to cool. The resultant mixture was partitioned between water (10 mL) and $CHCl_3$ (10 mL). The phases were separated then the organic phase was evaporated under a stream of nitrogen. The crude material was purified by flash column chromatography on a silica column (24 g). The column was eluted with a gradient of $CH_2Cl_2$:MeOH:$NH_4OH$ which was increased linearly from 99:1:0.1 to 95:5:0.5 over 10 CVs. The desired fractions were combined and evaporated to an orange gum (15 mg). The material thus obtained was purified further by flash column chromatography on a C-18 column (13 g). The column was eluted with a gradient of $H_2O$:MeCN (+0.1% $CF_3CO_2H$) which was increased linearly from 95:5 to 5:95 over 20 CVs. The desired fractions were combined and evaporated then partitioned between $CHCl_3$ (5 mL) and saturated aq. $NaHCO_3$ (5 mL). The organic phase was collected by passing through a hydrophobic frit then evaporated to yield the product as a red/brown gum (5 mg, 2.3%); $R_f$ 0.45 ($CH_2Cl_2$:MeOH:$NH_4OH$, 90:10:1); $\nu_{max}$ (neat) 2957 (C-H), 2929 (C-H), 2845 (C-H), 2808 (C-H), 2760 (C-H), 1650 (C=O); $^1$H NMR (500 MHz, $CDCl_3$) $\delta$ ppm 2.35 (s, 3 H, N$H_3$), 2.51 - 2.57 (m, 4 H, C(3)$H_2$+C(5)$H_2$), 3.33 - 3.40 (m, 4 H, C(2)$H_2$+C(6)$H_2$), 6.50 (d, *J*=2.5 Hz, 1 H, C(14)*H*), 6.87 (dd, *J*=8.0, 2.5 Hz, 1 H, C(12)*H*), 6.91 (dd, *J*=7.0, 0.5 Hz, 1 H, C(20)*H*), 7.30 (dd, *J*=9.0, 7.0 Hz, 1 H, C(19)*H*), 7.52 (s, 1 H, C(25)*H*), 7.66 - 7.71 (m, 2 H, C(18)*H*+C(24)*H*), 7.83 - 7.87 (m, 1 H, C(8)*H*), 9.78 (d, *J*=8.0 Hz, 1 H, C(11)*H*); $^{13}$C NMR (126 MHz, $CDCl_3$) $\delta$ ppm 46.0 (s, 1 C, N*C*$H_3$), 46.7 (s, 2 C, *C*(2)+*C*(6)), 54.2 (s, 2 C, *C*(3)+*C*(5)), 96.6 (s, 1 C, *C*(14)), 106.6 (s, 1 C, *C*(12)), 109.7 (s, 1 C, *C*(7)), 111.5 (s, 1 C, *C*(25)), 113.5 (s, 1 C, *C*(20)), 117.9 (q, *J*=289.5 Hz, 1 C, *C*$F_3$) 116.5 (s, 1 C, *C*(9)), 116.6 (s, 1 C, *C*(18)), 124.4 (s, 1 C, *C*(19)), 127.0 (q, *J*=3.5 Hz, 1 C, *C*(8)), 131.0 (s, 1 C, *C*(11)), 131.5 (s, 1 C, *C*(21)), 133.5 (s, 1 C, *C*(24)), 141.5 (s, 1 C, *C*(15)), 146.2 (s, 1 C, *C*(17)), 150.0 (s, 1 C, *C*(13)), 165.8 (q, *J*=35 Hz, 1 C, *C*(16)); LRMS *m/z* (ESI$^+$) 428 [MH$^+$]; HRMS (ESI$^+$) found 428.1697, calculated for $C_{22}H_{21}F_3N_5O$ 428.1693; HPLC (System D) $t_r$ 8.7 min (96%).

## Protein Expression and Purification

cDNA encoding reported human bromodomains were cloned, expressed and purified as previously described.[5,6]

## Differential Scanning Fluorimetry (DSF)

Thermal melting experiments were carried out using an Mx3005p Real Time PCR machine (Stratagene). Proteins were buffered in 10 mM HEPES pH 7.5, 500 mM NaCl and assayed in a 96-well plate at a final concentration of 2 µM in 20 µL volume. Compounds were added at a final concentration of 10 µM. SYPRO Orange (Molecular Probes) was used as a fluorescence probe at a dilution of 1:1000. Excitation and emission

filters for the SYPRO-Orange dye were set to 465 nm and 590 nm, respectively. The temperature was raised with a step of 3 °C per minute from 25 °C to 96 °C and fluorescence readings were taken at each interval. Data was analysed as previously described.[7]

## Isothermal Titration Calorimetry (ITC)

Experiments were carried out on a VP-ITC microcalorimeter (MicroCal™). All experiments were performed at 15 °C in 20 mM HEPES pH 7.5, 150 mM NaCl, 0.5 mM TCEP. BRD9 protein solution was buffer exchanged by gel filtration or dialysis into the ITC buffer. The titrations were conducted using an initial injection of 2 µl followed by 34 identical injections of 8 µl. The dilution heats were measured on separate experiments and were subtracted from the titration data. Thermodynamic parameters were calculated using $\Delta G = \Delta H - T\Delta S = -RT\ln K_B$, where $\Delta G$, $\Delta H$ and $\Delta S$ are the changes in free energy, enthalpy and entropy of binding respectively. In all cases a single binding site model was employed.

## Crystallization

BRD9 construct (Uniprot identifier as BRD9 HUMAN Q9H8M2-1 fragment 14-134) was used for all crystallographic studies. Aliquots of the purified proteins were set up for crystallization using a mosquito® crystallization robot (TTP Labtech). Coarse screens were typically setup onto Greiner 3-well plates using three different drop ratios of precipitant to protein per condition (200 + 100 nL, 150 + 150 nL and 100 + 200 nL). All crystallizations were carried out using the sitting drop vapour diffusion method at 4°C. BRD9 crystals with compound **28** (2 mM final concentration) were obtained by mixing 200 nL of the protein (14 mg/ml) and 100 nL crystallization buffer (0.2 M NaNO3, 0.1 M BTProp pH 6.5, 20% PEG 3350, 10% EtGly).

## Fluorescence Recovery After Photobleaching (FRAP) Assay

FRAP studies were performed using U2OS cells expressing a full-length BRD9 protein fused with an N-terminal eGFP as previously described.[8] In short, six hours after transfection 2.5 µM SAHA and compound **28** were added. Imaging was carried out 24 hours after transfection.

## Data Collection and Structure Solution

Crystals were cryo-protected using the well solution supplemented with additional 15% ethylene glycol and were flash frozen in liquid nitrogen. Data were collected at Diamond Light Source beamline I04-1 at a wavelength of 0.92 Å. Indexing and integration was carried out using XDS[9] and scaling was performed with AIMLESS.[10] Initial phases were calculated by molecular replacement with PHASER[11] using an ensemble of known bromodomain models (PDB IDs 2OSS, 2OUO, 2GRC, 2OO1, 3DAI, 3D7C, 3DWY, 3G0L). Unique and initial solutions were improved in a total of 50 cycles of automated protein chain tracing starting from existing model and computed using ARP/wARP. Further manual building with COOT and refinement against maximum likelihood target using REFMAC5. Thermal motions were analyzed using TLSMD[12] and hydrogen atoms were included in late refinement cycles. PRODRG[13] was used to generate compound coordinates and cif files. All model validations were carried out using MolProbity[14] Data collection and refinement statistics are compiled in Supplemental Table 1. The models and structure factors have been deposited into the pdb.

## Supplemental Table 1.

**BRD9 crystallographic data collection and refinement statistics.**

| | Ligand | **Compound** 28 |
|---|---|---|
| | **PDB ID** | XXXX |
| **Data Collection** | Space Group | P $2_1$ |
| Cell Dimensions | a,b,c (Å) | 58.38 37.96 60.55 |
| | α, β, γ (°) | 90.00 106.20 90.00 |
| | Resolution (Å) | 29.66 (1.80)* |
| | Unique Observations | 23,830 (1,368) |
| | Completeness (%) | 99.4 (96.2) |
| | Redundancy | 6.6 (6.3) |
| | $R_{sym}$ or $R_{merge}$ | 0.065 (0.657) |
| | $I/\sigma I$ | 19.5 (2.9) |
| | Wavelength | 0.9200 |
| | Phasing | MR |
| **Refinement** | $R_{work}/R_{free}$ (%) | 19.32 / 23.09 |
| Number of atoms | protein / other / solvent | 1840 / 62 / 136 |
| B-Factors (Å$^2$) | protein / other / solvent | 27.12 / 32.15 / 31.29 |
| | R.M.S.D. Bond (Å) | 0.016 |
| | R.M.S.D. Angle (°) | 1.574 |
| Ramachandran | Allowed (%) | 100.00 |
| statistics | Favored (%) | 100.00 |
| | Outliers (%) | 0.00 |

*Highest resolution shell (in Å) shown in parentheses

## Supplemental Table 2.

**Dissociation constants and thermodynamic parameters from BRD7 and BRD9 ITC assays for compound 28.**

| | **N**[1] | **ΔH (kcal/mol)** | **-TΔS (kcal/mol)**[2] | **ΔG (kcal/mol)** | **$K_a$ ($10^6$ $M^{-1}$)** |
|---|---|---|---|---|---|
| BRD9 | 0.956 ± 0.00321 | -10.7 ± 0.0664 | 1.32 | -9.38 | 14.8 ± 1.15 |
| BRD7 | 1.06 ± 0.00825 | -2.64 ± 0.0283 | -5.99 | -8.63 | 2.72 ± 0.200 |

[1] Molar binding ratio of the ligand-protein interaction (observed stoichiometry)
[2] At T = 298.15 K

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