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Synthesis of novel 3,4-diaryl-5-aminopyrazoles as potential kinase inhibitors

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ABSTRACT

Synthesis of a diverse series of novel 3,4-diaryl-5-aminopyrazoles as candidates in the development of new protein kinase inhibitors is reported for the first time. In the course of a wider study into bisindolylmaleimide (BIM) derivatives, we examined a novel 5-aminopyrazole heterocyclic moiety as a structural analogue of the highly potent VEGF-R2/3 inhibitor pyrrole-2-one (**8**). The versatile nature of this pharmacophore allows considerable scope for derivatisation and hence exploration of structure activity relationships. Consequently, a variety of structural modifications were used in order to diversify the aminopyrazole ring substituents. Bicyclic derivatives of the parent aminopyrazoles (**11**, **12**) were also synthesised as a means of probing the kinase active site, leading to formation of complex planar pyrimidine moieties. This work provides the framework for new explorations into kinase inhibition and critical investigations into selectivity of inhibitory activity.

1. Introduction

Protein kinases (PKs) constitute 1.7% of the total number of human genes and are one of the largest and most functionally disparate gene families in nature.[1] These enzymes control the synchronised function of downstream proteins by regulatory phosphorylation, orchestrating the flow of information from extracellular stimuli to intracellular signalling pathways.[2,3] As a validated target class for new mechanism-based therapeutic intervention, small-molecule inhibitors of aberrant kinase activity in epigenetic growth pathways constitute a central tenet of anti-cancer chemotherapy with low systemic toxicity.[4] Inhibition of deregulated kinases have also been indicated in treatment of diabetes (PKC-β), rheumatoid arthritis (p38 MAPK), and Parkinson's disease (MLK).[4–8] A small number of ATP-competitive small-molecule kinase inhibitors, such as Gleevec®, with a variety of selectivity profiles have now reached full clinical fruition; crystallographic data suggests a multitude of chemical scaffolds possess avid non-specific binding affinity for key residues occupying the widely conserved ATP-binding site, formed at the cleft between the N and C-terminal lobes.[9–11] Staurosporine **1** was identified as a pankinase inhibitor, competing for the ATP-binding site and exhibiting low nanomolar activity against PKC (IC_{50}=2.7 nM) and CDK1 (IC_{50}=4 nM).[5,12–14]

Structurally related to the unselective lead compound **1**, a number of 3,4-diarylmaleimides demonstrate relatively selective anti-tumour activity derived from inhibition within the aberrant kinome.[15] The binding mode of bisindolylmaleimides is more complex than compounds possessing a planar ring system, due to increased conformational distortion and hence, unique enzyme-ligand binding interactions.[16] Bisindolylmaleimide **2** was found to have an IC_{50} of 0.11 μM for PKC, implicated in unabated growth responses reported in several malignancies in vivo.[17] Replacement of an indole ring with a benzofuran moiety in **3** has been reported to afford selective and potent GSK-3β inhibition (Fig. 1).[18]

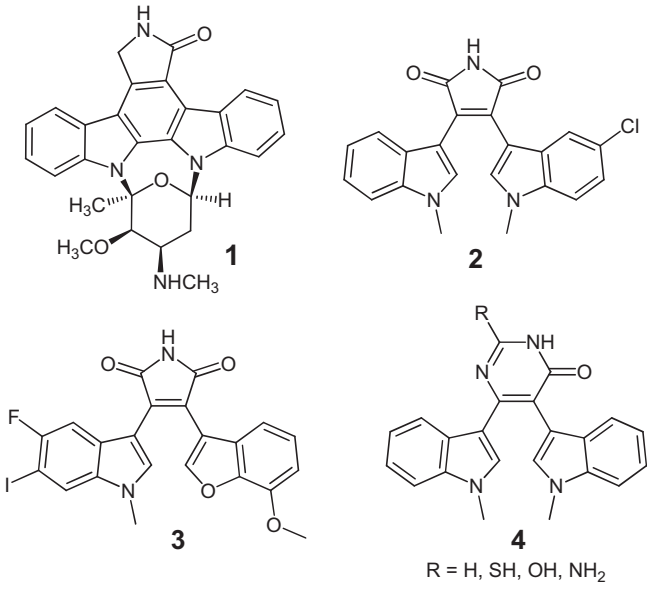

Fig. 1. Structures of small-molecule kinase inhibitors comprising a panel of modified diaryl substituted lactam derivatives.

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In this context, the potential of new flexible 3,4-diarylpyrrole congeners to yield synergistic kinase selectivity is a tantalising phenomenon at the forefront of current anti-cancer research. This report will thus investigate further structural insights into the key 'heterocyclic paradigm' of novel H-bonding anti-kinase inhibitor design, complementing our previously disclosed innovation of a library of bisindolylpyrimidin-4-ones **4**, currently being evaluated for in vitro anti-cancer activity.[19]

Treatment of numerous diseases involving dysregulation of vascular perfusion has also attracted pharmaceutical attention due to the remarkable success of inhibitors of VEGFR2 to disrupt pathologic tumour neovascularisation (solid tumour growth), age-related macular degeneration, rheumatoid arthritis and psoriasis.[20] An important contribution in this area was proffered by Peifer and co-workers following anti-angiogenesis evaluation of a panel of combretastatin A-4 analogues with a conserved stilbene moiety represented by the lead compound **5**. Rationalising that other *cisoid* 3,4-diaryl substituted compounds, such as **6** (containing a critical enzyme-interacting maleimide warhead) would display significant activity, preliminary compound screening confirmed 4-(3,4,5-trimethoxyphenyl)-3-(indol-3-yl)-1*H*-pyrrole-2,5-dione derivative **7** with an IC_{50} value of 2.5 nM for VEGFR2 (Fig. 2).[15]

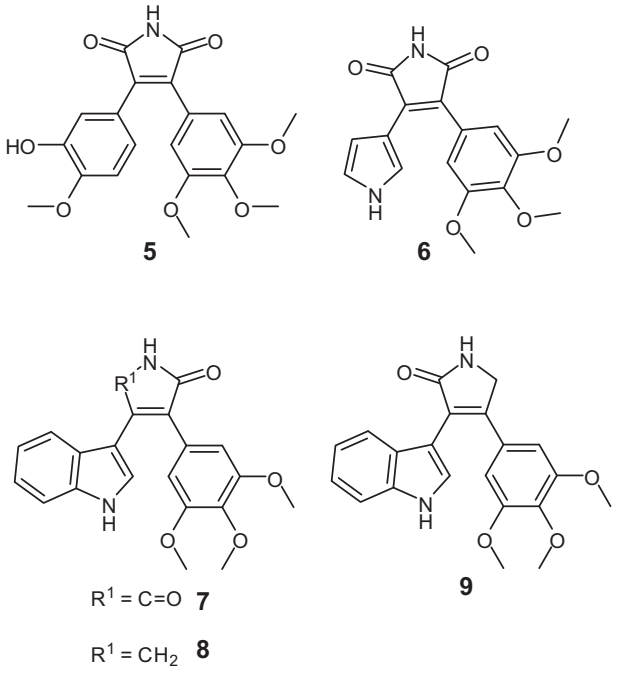


Fig. 2. Novel diaryl-substituted *N*-heterocyclic anti-kinase molecular templates conserving binary H-bonding amide network.

Envisaging that this aromatic substitution pattern comprised a key molecular scaffold, a simplified pyrrol-2-one analogue **8** was subsequently uncovered to exhibit attractive VEGFR2 inhibitory activity (IC_{50}=31 nM).[21] Modelling of **8** within the ATP site of VEGFR2 illustrated important protein–ligand interactions between the peptide backbone carbonyl of Glu915 and pyrrole NH, as well as between the backbone NH of Cys917 and hydrogen bond acceptor carbonyl group.[22] Introduction of enzyme selectivity aligned with strategic modification of this heterocyclic ring system was confirmed by the differential activity exhibited by regioisomeric pyrrolone **9** (IC_{50}=11 μM), hypothesised to result from altered orientation of the lactam in the kinase pocket (Fig. 2). Furthermore, this analogue **9** displayed single digit micromolar activity against other therapeutically-relevant kinases such as ARK5, Aurora A/B and SAK.[21] These data also illustrate an opportunity to develop unique activity profiles via application of novel inhibitor classes maintaining both active trimethoxyphenyl and indolyl substituents in place, while replacing the variable pyrrolic head-group present for compounds **7**–**9**. An example of this would represent an adaptable bidentate pyrrolic nucleus containing auxiliary polar character in order to exploit putative new modes of inhibitor binding within the ATP-binding kinase hinge region.

Recently, Bossi and co-workers revealed that PHA-E429 **10** (ALK; IC_{50}=91 nM) furnished an effective donor–acceptor–donor series of hydrogen bonds between the 5-aminopyrazole moiety and ALK hinge residues (Fig. 3).[23] Thus, comparison of rational binding modes for a 3-aryl-4-(indol-3-yl)-pyrrol-2-one scaffold with a closely analogous pyrazolic H-bonding orientation led us to identification of 3-(3,4,5-trimethoxyphenyl)-4-(indol-3-yl)-1*H*-pyrazole-5-amine derivatives containing a novel 5-aminopyrazole bioisostere as a potential new PK inhibitor class (**11**, Fig. 3). We now report synthesis of a library of novel derivatives based on the postulated ability of the diaryl-5-aminopyrazole system to mimic an active lactam conformation, as well as promote further H-bonding interactions in the kinase pocket.

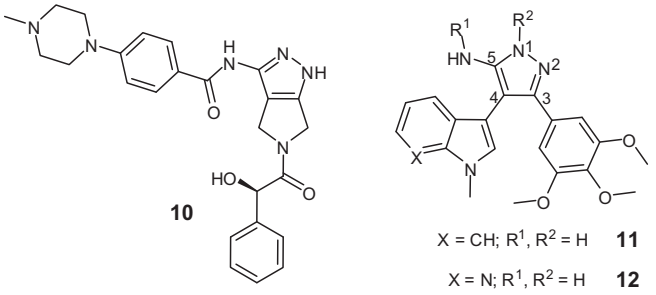


Fig. 3. Structures of PHA-E429 **10** containing H-bonding acylaminopyrazole and substituted 4-(indol-3-yl)-3-(3,4,5-trimethoxyphenyl)-5-aminopyrazole core (**11** and **12**).

It has been noted in the literature that replacement of an indole unit with a 7-azaindole moiety can alter inherent pharmacological properties in these compounds, while also enhancing drug-complex affinity by auxiliary formation of hydrogen bonds orientated within the active site of target enzymes.[24] In recent years, replacement of indole with 7-azaindole subunits in indolocarbazole and BIM analogues has consequently led to an awareness of selective cytotoxicity profiles associated with these congeners in vitro.[25,26] A complete series of 7-azaindole analogues (**12**; Fig. 3) was developed during this work, representing a key strategy for study of a 3-(7-azaindol-3-yl)-4-aryl-fused heterocyclic kinase inhibitory template.

In addition, the synthesis of complex purine-like pyrazolopyrimidine derivatives **13** and **14**, structurally related to 4,5-diaryl substituted pyrazolo[3,4-*c*]pyridazine **15**, a selective inhibitor of CDK1/cyclin B (IC_{50}=6.1 μM) (Fig. 4) has also been accomplished.[27]

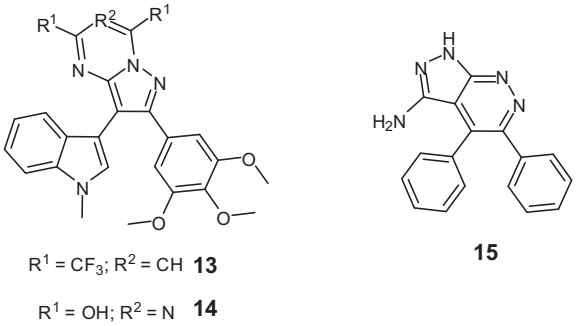


Fig. 4. Comparison of 4,5-diarylpyrazolo[3,4-*c*]pyridazine **15** with novel analogues comprising a fused bicyclic pharmacophore (**13**, **14**).

In this report, we describe the synthesis for the first time of these diaryl-5-aminopyrazoles, branched mono-*N*-substituted derivatives and bicyclic congeners.

2. Results and discussion

Previous reports describing the synthesis of 3-aryl-4-(indol-3-yl)pyrrole derivatives involve efficient application of Faul's Perkin-type methodology involving one-pot condensation between ethyl indole-3-glyoxylate and 3,4,5-trimethoxyphenylacetamide to afford 4-(3,4,5-trimethoxyphenyl)-3-(indol-3-yl)-1*H*-pyrrole-2,5-dione **7**, in 54% yield.[15] In 2008, the same group published their synthesis of reduced lactam **8**, by modified Knoevenagel route to the corresponding pyrrole-2-one, isolated following SEM-deprotection.[21]

In this work we present the targeted replacement of the pyrrol-2-one nucleus within compounds **8** and **9** resulting in formation of substituted 4-(1*H*-indol-3-yl)-3-(3,4,5-trimethoxyphenyl)-1*H*-pyrazol-5-amines **11** and **12**, via late-stage acid-promoted cyclocondensation in the presence of hydrazine.[28,29] Efforts to probe the molecular space within the inhibitor binding pocket were progressed by incorporation of functionality leading to elaboration of this key heterocyclic template.

2.1. 3,4-Diaryl-5-aminopyrazole synthesis (11)

Applying standard Mannich conditions starting from indole **16**, gramine **17** was initially isolated and converted to indole-3-acetonitrile **18**. *N*-Methylation was carried out following a literature procedure, to afford protected **19** in quantitative yield and used without purification.[30] Reaction of precursor **19** with LDA according to our previously reported modified Claisen conditions, with 3,4,5-trimethoxybenzoyl chloride **20**, as the product of acid chloride formation from acid **21**, afforded β-ketonitrile **22** in a yield of 35%. Following acetic acid-catalysed condensation of intermediate **22** with hydrazine, the polar 4-(1-methyl-1*H*-indol-3-yl)-3-(3,4,5-trimethoxyphenyl)-1*H*-pyrazol-5-amine **11** was isolated in a moderate yield of 23%, further diminished to 15% following subsequent scale-up. Fortunately, portionwise addition of excess hydrazine, along with camphoric acid in dry methanol at reflux for 24 h provided multi-gram synthesis of title compound **11** in a yield of 62% (Scheme 1).[31]

2.2. Derivatisation study

To define the versatility of our approach, an initial comparative library of 3-(3,4,5-trimethoxyphenyl)-4-(1-methyl-1*H*-indol-3-yl)-1*H*-pyrazol-5-amine analogues (**11**; $R^1/R^2 \neq H$) was successfully synthesised by suitable methodology, engineering a panel of novel compounds displaying unique hydrogen bonding networks.

Initial N^1-acetylation was accomplished following treatment with acetic anhydride at reflux overnight. On work-up, N^1-acetylated aminopyrazole **23** was purified by chromatography, and isolated as an off-white crystalline solid in a yield of 40%. Application of these conditions in the presence of trifluoroacetic anhydride provided the trifluoroacetamide analogue **24** (40%) (Scheme 2).

Construction of more complex derivatives bearing substituent H-bonding functionalities capable of exploiting side-chain interactions within the kinase hinge region also led to development of thioamide congener **25**, formed by heating in the presence of methyl isothiocyanate overnight, in an overall yield of 34% (Scheme 2).

Bioactive molecules bearing methylsulfonamide character also occupy an important role in current pharmaceutical research. Thus, reaction of aminopyrazole **11** with methanesulfonyl chloride in pyridine, at 90 °C, afforded compound **26** in a yield of 35%.

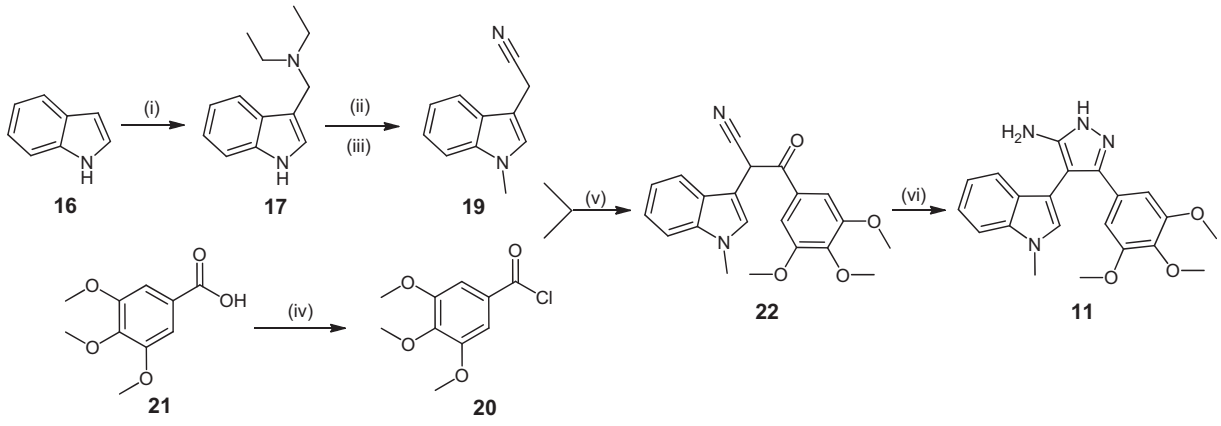
Synthesis of a novel N^1-guanylated compound **29** further expanded our scope to investigate congeners with attractive structural and electronic properties in this series. Base-promoted diimide coupling of **11** with (*N*,*N*′-di-*tert*-butoxycarbonyl)thiourea, **27**, initially gave access to the Boc-protected compound **28**, prior to acidic liberation of trifluoroacetate salt **29** in an overall yield of 35% (Scheme 2).[32]

2.3. Condensed bicyclic ring analogue

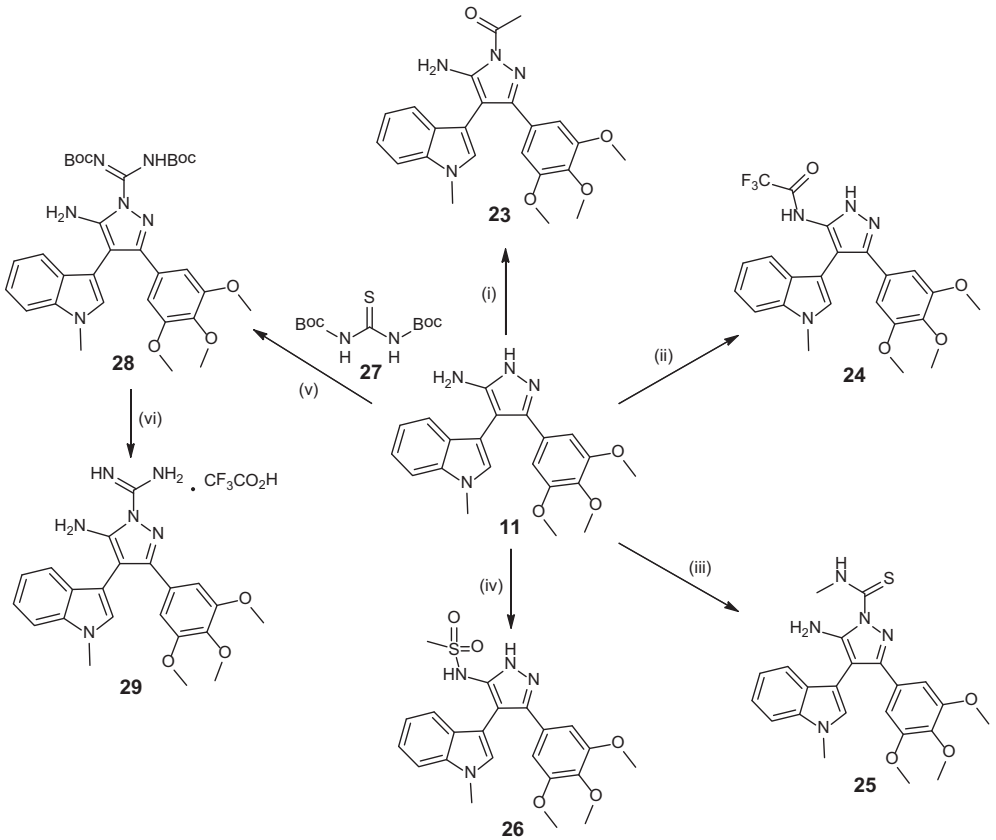
Formation of pyrazolo[1,5-*a*]pyrimidine compound **13** containing a planar pyrimidine pharmacophore was accomplished in 45% yield via acid-promoted condensation with HFAA, which was accompanied by rapid appearance of a persistent dark red colour within the reaction mixture. The final reaction in this derivatisation study was undertaken to form pyrazolotriazinedione **14**, incorporating a novel planar H-bonding scaffold comprising an attractive structural motif.[33] Formation of **14** under mild conditions proceeded in an excellent yield of 64%, requiring no further purification (Scheme 3).

2.4. 7-Azaindole analogue—β-ketonitrile intermediate and aminopyrazole nucleus

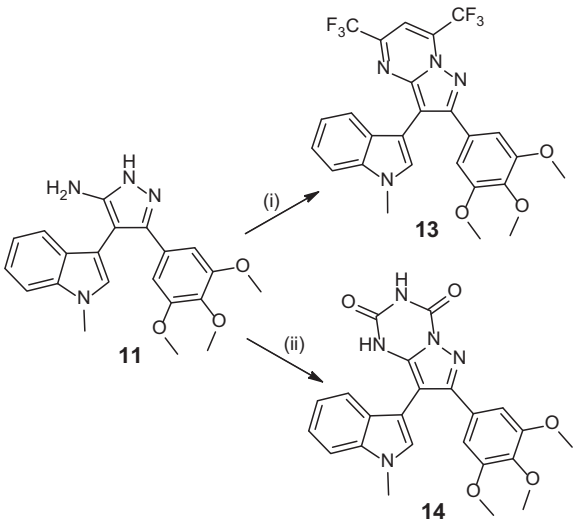
Initially, 7-azagramine **31** was synthesised via a Mannich reaction from 7-azaindole **30** using dimethylamine and formaldehyde (Scheme 4). Conversion to the corresponding acetonitrile **32** was achieved by displacement of the protonated amine using sodium cyanide. Interestingly, a novel diethylamine analogue **33** of 7-



Scheme 1. Reagents and conditions: (i) Et2NH, HCHO, MeOH, rt, 3 h, **17**=62%; (ii) CH_3I, KCN, MeOH, reflux, 20 h, **18**=67%; (iii) NaH, CH_3I, DMF, 0 °C to rt, 3 h, **19**=100%; (iv) $SOCl_2$, DCM, reflux, 20 h, **20**=100%; (v) (a) LDA, 3,4,5-$(OMe)_3C_6H_2COCl$ (**20**), THF, −78 °C to rt, 16 h (b) NH_4Cl/H_2O, **22**=35%; (vi) $NH_2NH_2.H_2O$, camphoric acid, MeOH, reflux, 24 h, **11**=62%.



Scheme 2. Reagents and conditions: (i) Ac_2O, CH_3CN, reflux, 16 h, **23**=40%; (ii) $(CF_3CO)_2O$, CH_3CN, reflux, 16 h, **24**=40%; (iii) (a) CH_3NCS, CH_3CN, reflux, 16 h (b) H_2O, 0 °C, 1 h, **25**=34%; (iv) CH_3SO_2Cl, C_6H_5N, 90 °C, 24 h, **26**=35%; (v) $HgCl_2$, **27**, Et_3N, DCM, 0 °C to rt, 16 h, **28**=35% (vi) TFA/DCM (10% v/v), 30 °C, 12 h, **29**=quant.



Scheme 3. Reagents and conditions: (i) HFAA, AcOH, 100 °C, 20 h, **13**=45%; (ii) (a) ClCONCO, Et_3N (2–3 drops), DCM, 0 °C to rt, 12 h (b) H_2O, 0 °C, 0.75 h, **14**=64%.

azagramine **31** had also been synthesised as a parallel to **17**, but all attempts to convert this precursor to acetonitrile **32** failed. Consequent protection of the indolic nitrogen was afforded using sodium hydride and methyl iodide. However, the yield of the desired product **34** was relatively low due to the formation of a dimethylated by-product, formed as a consequence of the highly acidic nature of the α-protons to the nitrile in this case.

Base-catalysed condensation of the *N*-protected acetonitrile **34** with the acid chloride **20** generated from **21** as previous, gave formation of β-ketonitrile intermediate **35**. Reaction of hydrazine with this key intermediate then afforded the parent aminopyrazole aza-analogue **12** in good yield, from which a series of derivatives of biological interest could be prepared.

2.5. Aminopyrazole monosubstitution

Parent aminopyrazole **12** readily underwent monosubstitution utilising a variety of different electrophiles (Scheme 5). However, similar to the corresponding indole derivatives, the site of substitution varied depending on the electrophile applied in each route.

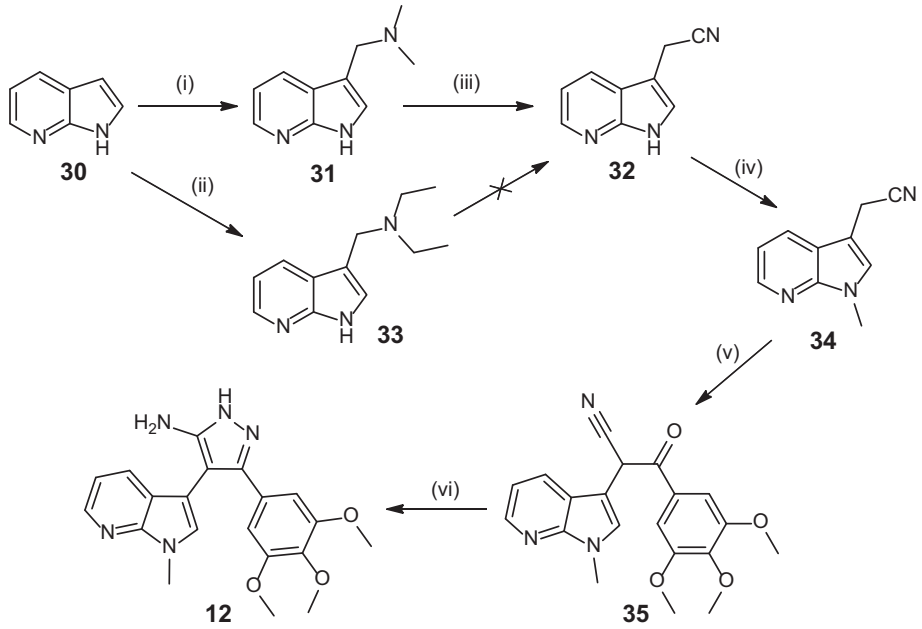
In the cases of methyl thiourea derivative **36** and acetyl derivative **37**, prepared by treatment of aminopyrazole **12** with methyl isothiocyanate and acetic anhydride, respectively, substitution occurred on the endocyclic nitrogen of the aminopyrazole ring. In the case of derivative **38**, prepared by treatment of aminopyrazole **12** with trifluoroacetic anhydride, trifluoroacetylation occurred on the exocyclic nitrogen of the ring, as confirmed by ^1H NMR spectroscopy (loss of the broad NH_2 peak between 4–6 ppm).

2.6. Preparation of bicyclic analogues

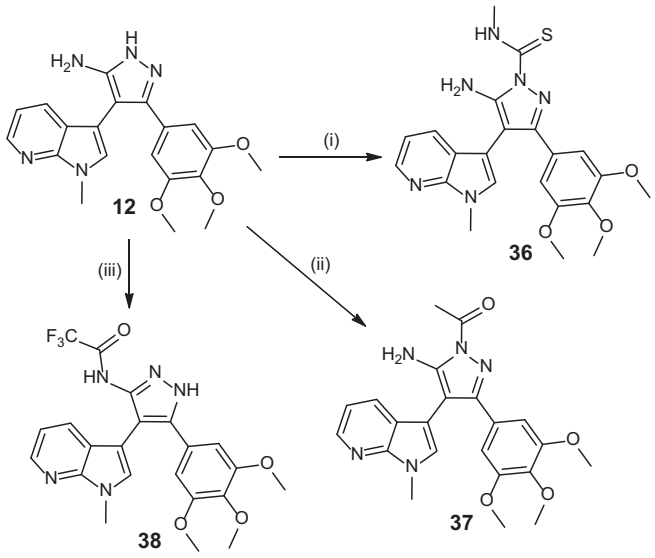
The triazinedione analogue **39** was synthesised following treatment of precursor **12** with *N*-chlorocarbonyl isocyanate (Scheme 6). A further bicyclic analogue of potential biological interest was synthesised, this time following condensation of **12** with HFAA, leading to the pyrimidine system **40**.

2.7. Regioselectivity of substitution

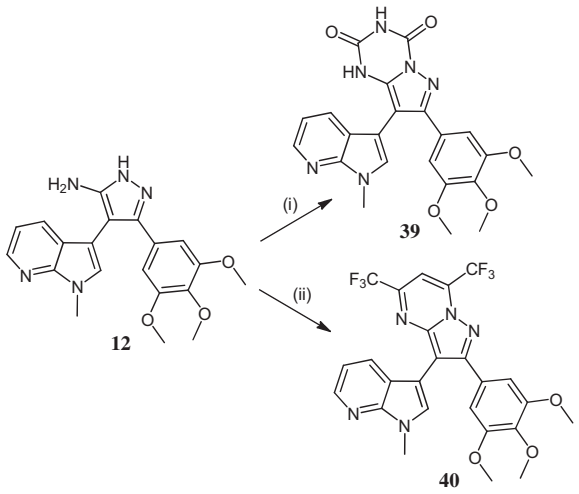
A recent comprehensive review of aminopyrazoles has postulated that regioselective substitution is heavily dependent on the



Scheme 4. Reagents and conditions: (i) $NH(CH_3)_2$, HCHO, AcOH, H_2O, EtOH, reflux, **31**=82% (ii) $NH(CH_2CH_3)_2$.HCl, HCHO, H_2O, EtOH, reflux, **33**=88% (iii) NaCN, AcOH, DMF, H_2O, reflux, **32**=52% (iv) NaH, MeI, DMF, 0 °C, **34**=46% (v) **20**, LDA, THF, −78 °C, **35**=49% (vi) $NH_2NH_2.H_2O$, cat. HCl, EtOH, reflux, **12**=70%.



Scheme 5. Reagents and conditions: (i) CH_3NCS, CH_3CN, reflux, **36**=47% (ii) Ac_2O, CH_3CN, reflux, **37**=62% (iii) $(CF_3CO)_2O$, CH_3CN, reflux, **38**=41%.



Scheme 6. Reagents and conditions: (i) ClCONCO, NEt_3, DCM, 0 °C to rt, **39**=56% (ii) HFAA, AcOH, reflux, **40**=54%.

nature of the electrophilic reagent and derivatisation conditions.[34] The observed monosubstitution pattern of these derivatives under our methodology was investigated by means of ^{1}H NMR spectroscopy. Evidence of N-5 or N-1 substitution is given from the presence or absence of signals relating to the amine (2H, broad peak between 4.5 and 6.5 ppm) or the pyrazole N–H (1H, broad peak at 13 ppm) as well as confirmatory evidence from single crystal X-ray structure analysis (data not shown). Consequently it can be seen that the amine is functionalized in the case of the trifluoroacetamide and the methanesulfonamide, whereas in all other cases the ring nitrogen was substituted. Interestingly, in the case of methanesulfonylation, reaction of **11** with methanesulfonyl chloride in refluxing acetonitrile, containing Et_3N was unsuccessful, while it was possible to access amino-substituted analogue **26** in the presence of pyridine at 90 °C indicating that thermal conditions may have a role.

This regiochemical outcome of these reactions requires further investigation in order to establish a protocol for selective substitution and this will be evaluated in future combinatorial studies through carbamate protection strategy.

3. Conclusion

A diverse series of novel 3,4-diarylaminopyrazoles were synthesised and fully characterised from β-ketonitrile intermediates. Derivatisation of the aminopyrazole core provided both bicyclic and monosubstituted scaffolds giving significant scope with which to explore the SAR for potential protein kinase inhibition. Reaction with bidentate electrophiles also resulted in the formation of more complex biologically relevant pyrimidine derivatives. Interestingly, with regard to a series of monosubstitution reactions, the aminopyrazole nucleus displayed reagent dependent regioselectivity as has been seen with similar systems but requires further investigation. In general, substitution occurred predominantly on the pyrazole nitrogen, rather than on the exocyclic amine, with the

exception of the trifluoroacetyl and methanesulfonyl derivatives. The synthetic procedures reported here provide a significant advance in the development of new and more selective kinase inhibitors and full combinatorial synthesis of this template is underway. Preliminary biological screening of both compound arrays has commenced and the selectivity/activity profiles of these diverse inhibitory candidates will also be explored.

4. Experimental

4.1. General procedures

Melting points were measured on a Uni-Melt Thomas Hoover Capillary Melting Point apparatus and are uncorrected. Low resolution mass spectra were recorded on a Waters Micromass Quattro Micro mass spectrometer (Instrument number QAA1202) in electrospray ionization (ESI) positive and negative modes and a Waters Micromass LCT Premier (Instrument number KD160) was used for high resolution acquisitions. Infrared (IR) spectra were recorded as potassium bromide (KBr) discs on a Perkin–Elmer FT-IR Paragon 1000 or a Spectrum One FT-IR spectrophotometer. ^{1}H (300 MHz) and ^{13}C NMR (75 MHz) NMR were recorded on a Bruker Avance 300 NMR spectrometer unless otherwise stated. All spectra were recorded at 20 °C in deuterated chloroform ($CDCl_3$) with tetramethylsilane (TMS) as an internal standard unless otherwise stated. Chemical shifts (δ_H and δ_C) are reported in parts per million (ppm), relative to TMS and coupling constants are expressed in hertz (Hz). Splitting patterns in ^{1}H NMR spectra are designated as s (singlet), br s (broad singlet), d (doublet), dd (doublet of doublets), ddd (doublet of doublet of doublets) dt (doublet of triplets), t (triplet), q (quartet) and m (multiplet). Thin layer chromatography (TLC) was carried out on precoated silica gel plates (Merck 60 PF_{254}). Visualisation was achieved by UV light (254 nm), vanillin or potassium permanganate staining. Column chromatography was carried out using Kieselgel 60, 0.040–0.063 nm (Merck).

4.1.1. 3-Diethylaminomethyl-1H-indole (17)[35]. To a 250 mL 2-neck round-bottomed flask was charged 37% formaldehyde solution (7 mL, 86 mmol), 98% diethylamine (8.9 mL, 86 mmol), along with indole (10.0 g, 86 mmol), absolute ethanol (120 mL) and zinc chloride (17.5 g, 128 mmol). The resultant straw coloured slurry was then stirred at room temperature for 2 h. The reaction mixture was then poured into a mixture of ethyl acetate (150 mL) and water (150 mL). Following addition of aqueous NaOH solution (20%) to reach pH 9, extraction was performed with 3×150 mL volumes of ethyl acetate; the resultant organic layers were evaporated under reduced pressure, diluted with water (120 mL) and acidified with concd HCl to pH 3. The slurry thus formed was then filtered under vacuum; the mother liquor was basified to pH 9, with further aqueous NaOH solution (20%). The turbid mixture was then cooled to 0 °C, and the light orange precipitate **17** filtered and washed with water, prior to drying (10.6 g, 62%): mp 105–107 °C (lit.[35] 109–110 °C); R_f (30% ethyl acetate/hexane) 0.45; ν_{max}/cm^{-1} (KBr) 3051, 2971, 1619, 1543, 1500, 1452; δ_H (300 MHz, $CDCl_3$) 1.06–1.11 [6H, t, *J* 7.1, 2×NCH_2CH_3], 2.54–2.61 [4H, q, *J* 7.1, 2×NCH_2CH_3], 3.79 [2H, s, C–H_2], 7.05–7.06 [1H, d, *J* 2.3, C–H_2], 7.07–7.13 [1H, td, *J* 7.5, 1.3, C–H_5], 7.14–7.19 [1H, td, *J* 7.8, 1.4, C–H_6], 7.29–7.32 [1H, d, *J* 8.0, C–H_7], 7.72–7.74 [1H, d, *J* 7.8, C–H_4], 8.32 [1H, br s, NH]; δ_C (75 MHz, $CDCl_3$) 12.0 ($2CH_3$, 2×NCH_2CH_3), 46.7 ($2CH_2$, 2×NCH_2CH_3), 48.0 (CH_2, $CH_2N(C_2H_5)_2$), 111.0 (CH, aromatic CH), 113.6 (C, aromatic C), 119.3 (CH, aromatic CH), 119.5 (CH, aromatic CH), 121.8 (CH, aromatic CH), 123.5 (CH, aromatic CH), 128.1 (C, aromatic C), 136.3 (C, aromatic C); *m*/*z* (ES^+) 203.1 $[M+H]^+$, (100%).

*4.1.2. 1H-Indol-3-yl acetonitrile (**18**)*[36]. To a solution of *N*,*N*-diethyl-3-aminomethyl indole **17** (0.370 g, 1.83 mmol) in methanol (10 mL) was added a solution of potassium cyanide (0.245 g, 3.77 mmol) in water (2 mL). Iodomethane (0.3 mL, 4.79 mmol) was added dropwise and the reaction mixture was then stirred vigorously at room temperature for 16 h. Following evaporation of the solvent, the crude residue was dissolved in ethyl acetate (100 mL) and successively washed with saturated aqueous sodium bicarbonate solution (100 mL), water (3×100 mL) and brine (2×100 mL). Following chromatography employing ethyl acetate/hexane, the title compound **18** was dried, and isolated as a beige amorphous solid (0.191 g, 67%): mp 32–34 °C (lit.[36] 34–36 °C); R_f (30% ethyl acetate/hexane) 0.35; ν_{max}/cm^{-1} (KBr) 3412, 2252, 1621, 1458, 1420; δ_H (300 MHz, $CDCl_3$) 3.82 [2H, s, C–H_2], 7.15–7.21 [2H, m, C–$H_{2,5}$], 7.22–7.28 [1H, td, *J* 7.1, 1.1, C–H_6], 7.37–7.40 [1H, d, *J* 8.0, C–H_7], 7.57–7.60 [1H, d, *J* 7.8, C–H_4], 8.19 [1H, br s, NH]; δ_C (75 MHz, $CDCl_3$) 14.4 (CH_2, CH_2CN), 104.7 (C, aromatic C), 111.6 (CH, aromatic CH), 118.1 (CH, aromatic CH), 118.2 (C, aromatic C), 120.3 (CH, aromatic CH), 122.8 (CH, aromatic CH), 122.9 (CH, aromatic CH), 126.0 (C, CH_2CN), 136.3 (C, aromatic C); *m*/*z* (ES^+) 130.0 $[M+H-HCN]^+$, (100%).

*4.1.3. 1-Methyl-1H-indol-3-yl acetonitrile (**19**)*[37]. To a suspension of NaH (60% dispersion in mineral oil) (3.33 g, 83.3 mmol) in anhydrous DMF (25 mL) was added dropwise a solution of 3-indolylacetonitrile **18** (10.0 g, 64 mmol) in DMF (50 mL). After stirring for 30 min, at room temperature, the flask was then cooled to 0 °C. Iodomethane (6 mL, 95.8 mmol) was added dropwise in solution with DMF (30 mL). The reaction was then allowed to warm and was stirred at ambient temperature for 3 h. The reaction was then quenched by pouring it into a mixture of ethyl acetate (300 mL) and 5% aqueous HCl (400 mL). Following product extraction, the organic layer was washed with water (2×250 mL), brine (300 mL), dried over magnesium sulfate and filtered. The resultant solution was then evaporated thoroughly, under reduced pressure, at a water bath temperature of 90 °C, to yield the *N*-methyl acetonitrile **19** in quantitative yield (11.1 g, 100%) as an off-white crystalline material: mp 57–59 °C (lit.[37] 59–60 °C); R_f (30% ethyl acetate/hexane) 0.40; ν_{max}/cm^{-1} (KBr) 2954, 2247, 1657, 1553, 1464; δ_H (300 MHz, $CDCl_3$) 3.73 [3H, s, NCH_3], 3.78 [2H, s, C–H_2], 7.04 [1H, s, C–H_2], 7.14–7.19 [1H, td, *J* 8.0, 1.5, C–H_5], 7.23–7.38 [2H, m, C–$H_{6,7}$], 7.54–7.57 [1H, d, *J* 7.9, C–H_4]; δ_C (75 MHz, $CDCl_3$) 14.3 (CH_2, CH_2CN), 32.9 (CH_3, NCH_3), 103.0 (C, aromatic C), 109.6 (CH, aromatic CH), 118.2 (CH, aromatic CH), 119.7 (CH, aromatic CH), 122.4 (CH, aromatic CH), 126.2 (C, CH_2CN), 126.5 (C, aromatic C), 127.3 (CH, aromatic CH), 137.1 (C, aromatic C); *m*/*z* (ES^+) 171.1 $[M+H]^+$, (100%).

*4.1.4. 3,4,5-Trimethoxybenzoyl chloride (**20**).* To a mixture of 3,4,5-trimethoxybenzoic acid (8.00 g, 37.7 mmol) in $CHCl_3$ (30 mL) was added thionyl chloride (13.7 mL, 189 mmol) in a dropwise fashion. Once the addition was completed, the reaction mixture was heated to reflux for 4 h before being cooled back to room temperature. The solvent and excess thionyl chloride was removed under reduced pressure to yield the acyl chloride **20** in quantitative yield as a colourless oil, which was subsequently used without further purification: ν_{max}/cm^{-1} (NaCl) 2970, 2944, 1750, 1590, 1457, 1415, 1130, 1128.

*4.1.5. 2-(1-Methyl-1H-indol-3-yl)-3-oxo-3-(3,4,5-trimethoxyphenyl) propanenitrile (**22**).* LDA solution (2 M) (19.4 mL, 38.7 mmol) was added, under nitrogen, to a flask containing THF (30 mL), cooled to −78 °C, and equilibrated at this temperature for 35 min, with stirring. Nitrile **19** (3.01 g, 17.6 mmol) in THF (4 mL) was then added slowly to the reaction flask and maintained at −78 °C for 2 h. 3,4,5-Trimethoxybenzoyl chloride (6.01 g, 26.4 mmol) was then dissolved in THF (15 mL) and added dropwise to the reaction vessel at −78 °C. The reaction was allowed to warm to room temperature and stirred overnight. Following solvent evaporation, the crude residue was taken up in ethyl acetate (100 mL) and washed with saturated

aqueous ammonium chloride solution (2×150 mL). These were then combined and further extracted with ethyl acetate (2×75 mL); the aggregate ethyl acetate layers were then washed with water (3×150 mL) and brine (2×150 mL), dried over magnesium sulfate, filtered and evaporated in vacuo. Employing a chromatographic gradient from 15 to 30% ethyl acetate/hexane, the novel intermediate **22** was isolated as a dark orange crystalline solid (2.22 g, 35%): mp 73–76 °C; R_f (30% ethyl acetate/hexane) 0.46; ν_{max}/cm^{-1} (KBr) 3433, 2938, 1684, 1584, 1504, 1127; δ_H (300 MHz, $CDCl_3$) 3.75 [3H, s, NCH_3], 3.76 [6H, s, 2×*m*-OCH_3], 3.86 [3H, s, *p*-OCH_3], 5.82 [1H, s, CH], 7.15 [1H, s, C–H_2], 7.18–7.24 [1H, t, *J* 7.3, C–H_5], 7.21 [2H, s, C–$H_{2',6'}$], 7.26–7.31 [1H, t, *J* 6.8, C–H_6], 7.32–7.35 [1H, d, *J* 7.5, C–H_7], 7.71–7.74 [1H, d, *J* 7.8, C–H_4]; δ_C (75 MHz, $CDCl_3$) 33.1 (CH_3, NCH_3), 38.4 (CH, *C*HCN), 56.3 (2CH_3, 2×*m*-OCH_3), 61.0 (CH_3, *p*-OCH_3), 103.9 (C, aromatic C), 106.7 (2CH, 2×aromatic CH), 110.1 (CH, aromatic CH), 116.9 (C, aromatic C), 118.1 (CH, aromatic CH), 120.6 (CH, aromatic CH), 122.9 (CH, aromatic CH), 125.6 (C, CN), 128.4 (CH, aromatic CH), 128.6 (C, aromatic C), 137.1 (C, aromatic C), 143.4 (C, aromatic C), 153.0 (2C, 2×aromatic C), 187.5 (C, C=O); *m*/*z* (ES^+) 365.1 $[M+H]^+$, (60%); (ES^-) 363.1 $[M-H]^-$, (100%); HRMS (ES^+) exact mass calculated for $C_{21}H_{21}N_2O_4$ $[M+H]^+$ 365.1501. Found 365.1486.

*4.1.6. 4-(1-Methyl-1H-indol-3-yl)-3-(3,4,5-trimethoxyphenyl)-1H-pyrazol-5-amine (**11**).* Hydrazine hydrate (0.14 mL, 1.5 mmol) and camphoric acid (0.180 g, 0.88 mmol, 1.2 equiv) were added to a suspension of the β-ketonitrile **22** (0.265 g, 0.73 mmol), in dry methanol (10 mL). The reaction mixture was then stirred at reflux temperature for 3 h, prior to addition of excess hydrazine hydrate (0.42 mL, 4.5 mmol) and heating for a further 20 h. Following solvent removal, the residue was taken up in ethyl acetate (50 mL) and washed with saturated aqueous sodium bicarbonate solution (2×70 mL). These aqueous bicarbonate layers were added together and subsequently extracted with ethyl acetate (2×30 mL); the combined ethyl acetate layers were then washed with water (3×70 mL) and brine (2×70 mL), dried over magnesium sulfate, filtered and evaporated to yield a crude golden brown solid. Following purification by flash chromatography, initially with 100% ethyl acetate, to complete full elution of the starting material, and then with 98% ethyl acetate/2% methanol, the polar product **11** was isolated as light brown crystals (0.170 g, 62%): mp 117–120 °C; R_f (2% methanol/ethyl acetate) 0.20; ν_{max}/cm^{-1} (KBr) 2930, 1583, 1511, 1465, 1422, 1124; δ_H (300 MHz, $CDCl_3$) 3.42 [6H, s, 2×*m*-OCH_3], 3.77 [3H, s, NCH_3], 3.81 [3H, s, *p*-OCH_3], 4.75 [2H, br s, NH_2], 6.68 [2H, s, C–$H_{2',6'}$], 7.01–7.06 [3H, m, C–$H_{2,5}$ and NH], 7.20–7.25 [1H, t, *J* 8.0, C–H_6], 7.30–7.33 [1H, d, *J* 7.8, C–H_7], 7.33–7.36 [1H, d, *J* 8.2, C–H_4]; δ_C (75 MHz, $CDCl_3$) 32.9 (CH_3, NCH_3), 55.7 (2CH_3, 2×*m*-OCH_3), 60.8 (CH_3, *p*-OCH_3), 104.0 (2CH, 2×aromatic CH), 104.4 (C, aromatic C), 105.7 (C, aromatic C), 109.3 (CH, aromatic CH), 119.6 (CH, aromatic CH), 120.4 (CH, aromatic CH), 122.0 (CH, aromatic CH), 126.1 (C, aromatic C), 127.9 (C, aromatic C), 128.0 (C, aromatic C), 128.4 (CH, aromatic CH), 137.1 (C, aromatic C), 137.8 (C, aromatic C), 153.1 (C, aromatic C), 153.3 (2C, 2×aromatic C); *m*/*z* (ES^+) 379.1 $[M+H]^+$, (100%); HRMS (ES^+) exact mass calculated for $C_{21}H_{23}N_4O_3$ $[M+H]^+$ 379.1770. Found 379.1765.

*4.1.7. N-(4-(1-Methyl-1H-indol-3-yl)-3-(3,4,5-trimethoxyphenyl)-1H-pyrazol-5-yl) acetamide (**23**).* To a solution of aminopyrazole **11** (0.150 g, 0.397 mmol) in acetonitrile (10 mL) was added acetic anhydride (0.05 mL, 0.476 mmol) and the reaction mixture was heated to 80 °C, with stirring, for 16 h. Following solvent removal, the crude acetylated product was dissolved in ethyl acetate (30 mL), washed with 10% aqueous sodium bicarbonate solution (50 mL), followed by water (3×50 mL) and brine (50 mL). The organic layer was then dried over magnesium sulfate, prior to filtration and evaporation of the filtrate to a crude residue. Chromatography with 15% ethyl acetate/hexane, followed by concentration of the product fractions, generated the pure pyrazolic acetamide derivative **23** as off-white plate-like crystals (0.067 g, 40%): mp 182–184 °C; R_f (30% ethyl acetate/hexane) 0.71; ν_{max}/cm^{-1} (KBr) 3449, 3338, 2935, 1710, 1608, 1572, 1485, 1126; δ_H (300 MHz, $CDCl_3$) 2.79 [1H, s, $COCH_3$], 3.45 [6H, s, 2×*m*-OCH_3], 3.78 [3H, s, NCH_3], 3.82 [3H, s, *p*-OCH_3], 5.46 [2H, br s, NH_2], 6.87 [2H, s, C–$H_{2',6'}$], 6.99 [1H, s, C–H_2], 7.06–7.11 [1H, td, *J* 8.0, 0.9, C–H_5], 7.23–7.28 [1H, td, *J* 8.1, 1.1, C–H_6], 7.35–7.38 [1H, d, *J* 8.0, C–H_7], 7.38–7.40 [1H, d, *J* 7.6, C–H_4]; δ_C (75 MHz, $CDCl_3$) 23.2 (CH_3, $COCH_3$), 32.9 (CH_3, NCH_3), 55.6 (2CH_3, 2×*m*-OCH_3), 60.8 (CH_3, *p*-OCH_3), 94.5 (C, aromatic C), 104.7 (C, aromatic C), 105.0 (2CH, 2×aromatic CH), 109.4 (CH, aromatic CH), 119.8 (CH, aromatic CH), 120.2 (CH, aromatic CH), 122.2 (CH, aromatic CH), 127.9 (C, aromatic C), 128.0 (C, aromatic C), 128.7 (CH, aromatic CH), 137.2 (C, aromatic C), 138.4 (C, aromatic C), 148.7 (C, aromatic C), 152.8 (2C, 2×aromatic C), 153.3 (C, aromatic C), 173.8 (C, C=O); *m*/*z* (ES^+) 421.2 $[M+H]^+$, (100%); HRMS (ES^+) exact mass calculated for $C_{23}H_{25}N_4O_4$ $[M+H]^+$ 421.1876. Found 421.1862.

*4.1.8. 2,2,2-Trifluoro-N-(4-(1-methyl-1H-indol-3-yl)-3-(3,4,5-trimethoxyphenyl)-1H-pyrazol-5-yl) acetamide (**24**).* Trifluoroacetic anhydride (0.10 mL, 0.63 mmol) was added in a single portion, to a mixture of parent aminopyrazole **11** (0.200 g, 0.53 mmol) and acetonitrile (10 mL). The dark-brown reaction mixture was then heated to reflux for 16 h. The solvent was then evaporated in vacuo, to yield the trifluoroacetylated product **24** as a light orange solid, following column chromatography (30% ethyl acetate/hexane) of the crude residue (0.100 g, 40%): mp 113–116 °C; R_f (30% ethyl acetate/hexane) 0.69; ν_{max}/cm^{-1} (KBr) 3270, 2939, 1729, 1585, 1504, 1466, 1126; δ_H (300 MHz, $CDCl_3$) 3.43 [6H, s, 2×*m*-OCH_3], 3.78 [3H, s, NCH_3], 3.86 [3H, s, *p*-OCH_3], 6.75 [2H, s, C–$H_{2',6'}$], 7.05–7.11 [2H, m, C–$H_{5,6}$], 7.26 [1H, s, C–H_2], 7.26–7.31 [2H, t, *J* 7.0, C–H_7], 7.39–7.41 [1H, d, *J* 8.3, C–H_4], 8.17 [2H, br s, $NHCOCF_3$ and pyrazole NH]; δ_C (75 MHz, $CDCl_3$) 33.1 (CH_3, NCH_3), 55.6 (2CH_3, 2×*m*-OCH_3), 60.9 (CH_3, *p*-CH_3), 103.2 (C, aromatic C), 104.2 (2×CH, aromatic CH), 105.2 (CH, aromatic CH), 109.6 (CH, aromatic CH), 119.9 (CH, aromatic CH), 120.3 (CH, aromatic CH), 122.7 (CH, aromatic CH), 127.5 (C, aromatic C), 128.5 (2×C, aromatic C), 133.6 (C, aromatic C), 137.3 (2×C, aromatic C), 143.3 (C, aromatic C), 153.1 (2×C, aromatic C); *m*/*z* (ES^+) 475.1 $[M+H]^+$, (100%); HRMS (ES^+) exact mass calculated for $C_{23}H_{22}N_4O_4F_3$ $[M+H]^+$ 475.1593. Found 475.1591.

*4.1.9. 1-Methyl-3-[4-(1-methyl-1H-indol-3-yl)-3-(3,4,5-trimethoxyphenyl)-1H-pyrazol-5-yl] thiourea (**25**).* Methyl isothiocyanate (0.040 g, 0.55 mmol) was applied to a stirred solution of compound **11** (0.200 g, 0.53 mmol) in acetonitrile (10 mL). The reaction was then heated to reflux temperature, and the dark orange suspension was stirred for 16 h. The reaction was initially allowed to cool to room temperature, and subsequently, to 0 °C, and maintained for 30 min at this temperature. The formed slurry was then filtered to yield pure thiourea derivative **25** as light orange crystals (0.080 g, 34%): mp 186–188 °C; R_f (35% ethyl acetate/hexane) 0.80; ν_{max}/cm^{-1} (KBr) 3418, 3272, 2931, 1624, 1519, 1423, 1117; δ_H (300 MHz, $CDCl_3$) 3.28–3.30 [3H, d, *J* 4.9, NH*C*H_3], 3.43 [6H, s, 2×*m*-OCH_3], 3.78 [3H, s, NCH_3], 3.82 [3H, s, *p*-OCH_3], 6.35 [2H, s, NH_2], 6.85 [2H, s, C–$H_{2',6'}$], 7.01 [1H, s, C–H_2], 7.06–7.11 [1H, t, *J* 7.3, C–H_5], 7.23–7.28 [1H, t, *J* 8.9, C–H_6], 7.35–7.38 [1H, d, *J* 8.1, C–H_7], 7.38–7.40 [1H, d, *J* 7.8, C–H_4], 9.38 [1H, s, N*H*CH_3]; δ_C (75 MHz, $CDCl_3$) 29.0 (CH_3, NH*C*H_3), 31.1 (CH_3, NCH_3), 53.8 (2CH_3, 2×*m*-OCH_3), 59.0 (CH_3, *p*-OCH_3), 93.1 (C, aromatic C), 103.1 (C, aromatic C), 103.2 (2CH, 2×aromatic CH), 107.5 (CH, aromatic CH), 118.0 (CH, aromatic CH), 118.4 (CH, aromatic CH), 120.4 (CH, aromatic CH), 125.7 (C, aromatic C), 126.2 (C, aromatic C), 127.1 (CH, aromatic CH), 135.3 (C, aromatic C), 136.6 (C, aromatic C), 148.1 (C, aromatic C), 148.5 (C, aromatic C), 151.0 (2C, 2×aromatic C), 175.1 (C, C=S); *m*/*z*

(ES^+) 452.1 $[M+H]^+$, (50%); HRMS (ES^+) exact mass calculated for $C_{23}H_{26}N_5O_3S$ $[M+H]^+$ 452.1756. Found 452.1742.

*4.1.10. N-(4-(1-Methyl-1H-indol-3-yl)-3-(3,4,5-trimethoxyphenyl)-1H-pyrazol-5-yl) methanesulfonamide (**26**).* To a solution of aminopyrazole **11** (0.108 g, 0.29 mmol) in pyridine (8 mL) was added methanesulfonyl chloride (0.023 mL, 0.29 mmol), along with triethylamine (0.04 mL, 0.29 mmol). The reaction was heated at reflux overnight and then evaporated in vacuo. The dark residue was dissolved in ethyl acetate (80 mL), washed with 10% aqueous HCl (5×70 mL), saturated aqueous sodium bicarbonate solution (100 mL), water (3×100 mL) and brine (100 mL), dried over magnesium sulfate and evaporated under reduced pressure. Chromatography employing 25% ethyl acetate/hexane yielded the final product **26** as a dark brown solid (0.048 g, 35%): mp 129–132 °C; R_f (40% ethyl acetate/hexane) 0.55; ν_{max}/cm^{-1} (KBr) 3307, 2930, 1584, 1465, 1424, 1125; δ_H (300 MHz, $CDCl_3$) 3.24 [3H, s, SO_2CH_3], 3.42 [6H, s, 2×*m*-OCH_3], 3.78 [3H, s, NCH_3], 3.82 [3H, s, *p*-OCH_3], 6.61 [2H, s, C–$H_{2',6'}$], 7.00–7.05 [1H, t, *J* 7.4, C–H_5], 7.08 [1H, s, C–H_2], 7.17–7.19 [1H, d, *J* 7.8, C–H_7], 7.19–7.24 [1H, t, *J* 7.5, C–H_6], 7.26 [1H, s, pyrazole NH], 7.33–7.35 [1H, d, *J* 8.16, C–H_4]; δ_C (75 MHz, $CDCl_3$) 33.0 (CH_3, NCH_3), 41.4 (CH_3, SO_2CH_3), 55.8 (2CH_3, 2×*m*-OCH_3), 60.9 (CH_3, *p*-OCH_3), 103.4 (C, aromatic C), 103.5 (C, aromatic C), 104.0 (2CH, 2×aromatic CH), 109.5 (CH, aromatic CH), 119.9 (CH, aromatic CH), 120.0 (CH, aromatic CH), 122.3 (CH, aromatic CH), 124.6 (C, aromatic C), 127.6 (C, aromatic C), 128.9 (CH, aromatic CH), 137.1 (C, aromatic C), 138.3 (C, aromatic C), 141.8 (C, aromatic C), 145.2 (C, aromatic C), 153.3 (2C, 2×aromatic C); *m*/*z* (ES^+) 457.1 $[M+H]^+$, (100%); HRMS (ES^+) exact mass calculated for $C_{22}H_{25}N_4O_5S$ $[M+H]^+$ 457.1546. Found 457.1550.

*4.1.11. (N,N′-Di-tert-butoxycarbonyl)-thiourea (**27**)[38].* To a stirred solution of NaH (1.35 g, 33.8 mmol) (60% suspension in mineral oil) in dry THF (150 mL), under inert conditions at 0 °C, was added thiourea (0.570 g, 7.5 mmol) portionwise over 5 min, and left to stir at room temperature for 15 min. Following cooling to 0 °C, di-*tert*-butyl carbonate (3.60 g, 16.5 mmol) was added, and stirring was continued for 40 min, prior to warming to room temperature over 3 h. The reaction was subsequently quenched via the addition of saturated aqueous $NaHCO_3$ solution (10 mL), water (200 mL) and the product was extracted with ethyl acetate (3×75 mL). On drying with magnesium sulfate, the filtrate was evaporated to dryness to afford **27** as an off-white solid, used successfully without further purification (1.91 g, 92%): mp 115–117 °C (lit.[38] 120–122 °C); ν_{max}/cm^{-1} (KBr) 3365, 3173, 2925, 1771, 1724, 1609, 1542; δ_H (300 MHz, $CDCl_3$) 1.48 [9H, s, $C(CH_3)_3$], 1.53 [9H, s, $C(CH_3)_3$], 8.26 [1H, s, NH], 9.23 [1H, s, NH]; δ_C (75 MHz, $CDCl_3$) 28.0 (6CH_3, 2×$C(CH_3)_3$), 84.1 (2C, 2×$C(CH_3)_3$), 151.3 (C, C=S), 177.9 (C, C=O), 182.1 (C, C=O); *m*/*z* (ES^+) 157.0 $[M+H]^+$, (50%), (ES^-) 155.0 $[M-H]^+$, (45%).

*4.1.12. tert-Butyl (tert-butoxycarbonylamino) (4-(1-methyl-1H-indol-3-yl)-3-(3,4,5-trimethoxyphenyl)-1H-pyrazol-5-ylamino)methylenecarbamate (**28**).* Aminopyrazole **11** (0.150 g, 0.397 mmol) was treated with $HgCl_2$ (0.130 g, 0.437 mmol) and *N*,*N*′-di-*tert*-butoxycarbonyl thiourea **27** (0.109 g, 0.397 mmol), in distilled DCM (10 mL), at 0 °C. Triethylamine (0.2 mL, 1.23 mmol) was then added to the flask and allowed to stir at 0 °C for a further 1 h, followed by stirring for 12 h at room temperature. The reaction was then poured into ethyl acetate (30 mL), and filtered through a pad of Celite® in order to remove the yellow mercury sulfide by-product. The filter cake was washed with ethyl acetate (3×15 mL). The combined organic phases were then washed with water (3×100 mL) and brine (100 mL), dried over magnesium sulfate and concentrated under vacuum. Chromatography employing 15% ethyl acetate/hexane yielded the pure di-protected pyrazole derivative **28** as an off-white crystalline solid (0.085 g, 35%): mp 153–156 °C; R_f (25% ethyl acetate/hexane) 0.85; ν_{max}/cm^{-1} (KBr) 3487, 3360, 2975, 1766, 1751, 1712, 1656, 1587, 1126; δ_H (300 MHz, $CDCl_3$) 1.55 [9H, s, $CO_2C(CH_3)_3$], 1.56 [9H, s, $CO_2C(CH_3)_3$], 3.47 [6H, s, 2×*m*-OCH_3], 3.78 [3H, s, NCH_3], 3.82 [3H, s, *p*-OCH_3], 5.72 [2H, br s, NH_2], 6.81 [2H, s, C–$H_{2',6'}$], 6.98 [1H, s, C–H_2], 7.07–7.12 [1H, td, *J* 7.8, 0.7, C–H_5], 7.23–7.28 [1H, t, *J* 7.7, C–H_6], 7.35–7.38 [1H, d, *J* 8.3, C–H_7], 7.36–7.39 [1H, d, *J* 7.9, C–H_4], 9.25 [1H, s, N*H*$CO_2C(CH_3)_3$]; δ_C (75 MHz, $CDCl_3$) 28.1 (3CH_3, $CO_2C(CH_3)_3$), 28.2 (3CH_3, $CO_2C(CH_3)_3$), 32.9 (CH_3, NCH_3), 55.7 (2CH_3, 2×*m*-OCH_3), 60.8 (CH_3, *p*-OCH_3), 81.0 (C, $CO_2C(CH_3)_3$), 83.1 (C, $CO_2C(CH_3)_3$), 95.2 (C, aromatic C), 104.7 (C, aromatic C), 105.2 (2CH, 2×aromatic CH), 109.4 (CH, aromatic CH), 119.9 (CH, aromatic CH), 120.1 (CH, aromatic CH), 122.2 (CH, aromatic CH), 127.6 (C, aromatic C), 127.9 (C, aromatic C), 128.8 (CH, aromatic CH), 137.1 (C, aromatic C), 138.5 (C, aromatic C), 143.2 (C, aromatic C), 148.6 (C, aromatic C), 149.7 (C, NH-C=N), 152.0 (C, C=O), 152.9 (2C, 2×aromatic C), 157.3 (C, C=O); *m*/*z* (ES^+) 621.3 $[M+H]^+$, (100%); HRMS (ES^+) exact mass calculated for $C_{32}H_{41}N_6O_7$ $[M+H]^+$ 621.3037. Found 621.3033.

*4.1.13. 1-(4-(1-Methyl-1H-indol-3-yl)-3-(3,4,5-trimethoxyphenyl)-1H-pyrazol-5-yl)guanidinium trifluoroacetate (**29**).* Boc-protected derivative **28** (0.05 g, 0.08 mmol) was treated, at 30 °C, with a 10% v/v solution of trifluoroacetic acid in DCM (15 mL), for 12 h. After stirring for this time, the solvent was evaporated in vacuo to generate the corresponding trifluoroacetate salt **29** in quantitative yield: mp 118–120 °C; ν_{max}/cm^{-1} (KBr) 3369, 1698, 1551, 1421; *m*/*z* (ES^+) 421.2 $[M]^+$, (100%); (ES^-) 112.9 $[CF_3CO_2]^-$, (100%).

*4.1.14. 3-(1-Methyl-1H-indol-3-yl)-5,7-bis(trifluoromethyl)-2-(3,4,5-trimethoxyphenyl) pyrazolo[1,5-a]pyrimidine (**13**).* Hexafluoroacetylacetone (1.50 mL, 10.6 mmol) was added to a mixture of compound **11** (0.200 g, 0.53 mmol) and acetic acid (10 mL), with stirring, at room temperature. The reaction was then heated to 80 °C for 20 h, at which point, full consumption of the starting material had occurred. Following removal of acetic acid under vacuum, the crude hexafluoropyrimidine derivative **13** was purified by chromatography employing a gradient from 20 to 50% ethyl acetate/hexane. The fractions containing the product were then combined prior to being evaporated in vacuo, and then dried under high vacuum overnight, to yield pyrimidine **13** as a dark red crystalline solid (0.130 g, 45%): mp 217–220 °C; R_f (30% ethyl acetate/hexane) 0.65; ν_{max}/cm^{-1} (KBr) 2940, 1577, 1485, 1467, 1420, 1170; δ_H (300 MHz, $CDCl_3$) 3.48 [6H, s, 2×*m*-OCH_3], 3.84 [3H, s, NCH_3], 3.90 [3H, s, *p*-OCH_3], 6.97–7.02 [1H, t, *J* 7.2, C–H_5], 7.10–7.13 [1H, d, *J* 7.8, C–H_7], 7.12 [2H, s, C–$H_{2',6'}$], 7.19–7.25 [1H, t, *J* 7.2, C–H_6], 7.35 [1H, s, $(CF_3)C-CH=C(CF_3)$], 7.36–7.39 [1H, d, *J* 8.3, C–H_4], 7.42 [1H, s, C–H_2]; δ_C (75 MHz, $CDCl_3$) 33.1 (CH_3, NCH_3), 55.7 (2CH_3, 2×*m*-OCH_3), 60.9 (CH_3, *p*-OCH_3), 102.1 (CH, aromatic CH), 103.3 (C, aromatic C), 106.1 (2CH, 2×aromatic CH), 106.7 (C, aromatic C), 109.4 (CH, aromatic CH), 120.4 (C, q, J_{F-C} 274.3, CF_3), 121.7 (C, q, J_{F-C} 274.4, CF_3), 119.8 (CH, aromatic CH), 121.0 (CH, aromatic CH), 122.1 (CH, aromatic CH), 126.9 (C, aromatic C), 127.3 (C, aromatic C), 129.5 (CH, aromatic CH), 134.7 (C, q, J_{F-C} 38.6, aromatic C), 137.2 (C, aromatic C), 139.2 (C, aromatic C), 144.2 (C, q, J_{F-C} 37.8, aromatic C), 147.1 (C, aromatic C), 153.1 (2C, 2×aromatic C), 156.0 (C, aromatic C); *m*/*z* (ES^+) 551.2 $[M+H]^+$, (100%); HRMS (ES^+) exact mass calculated for $C_{26}H_{21}N_4O_3F_6$ $[M+H]^+$ 551.1518. Found 551.1514.

*4.1.15. 8-(1-Methyl-1H-indol-3-yl)-7-(3,4,5-trimethoxyphenyl)pyrazolo[1,5-a][1,3,5]triazine-2,4(1H,3H)-dione (**14**).* Chlorocarbonyl isocyanate (0.05 mL, 0.582 mmol) was added to a stirred solution of intermediate **11** (0.100 g, 0.265 mmol) in distilled DCM (5 mL), containing two drops of triethylamine, at 0 °C. The reaction was then allowed to stir at room temperature overnight; water (1 mL) was added carefully to the flask and some effervescence was observed. The resultant dark brown slurry was filtered and the

isolated brown solid was washed successively with water, ether and air-dried for 1 h, to afford the title compound **14** (0.075 g, 64%): mp 268–271 °C; R_f (30% ethyl acetate/hexane) 0.18; ν_{max}/cm^{-1} (KBr) 3433, 3213, 2939, 1763, 1710, 1587, 1419, 1125; δ_H (300 MHz, DMSO-d_6) 3.38 [6H, s, 2×*m*-OCH_3], 3.65 [3H, s, NCH_3], 3.90 [3H, s, *p*-OCH_3], 6.93 [2H, s, C–$H_{2',6'}$], 7.02–7.06 [1H, t, *J* 7.6, C–H_5], 7.17–7.19 [1H, d, *J* 7.8, C–H_7], 7.22–7.27 [3H, t, *J* 8.1, C–H_6], 7.48 [1H, s, C–H_2], 7.55–7.58 [1H, d, *J* 8.3, C–H_4], 11.67 [2H, br s, 2×NH]; δ_C (75 MHz, DMSO-d_6) 32.5 (CH_3, NCH_3), 55.1 ($2CH_3$, 2×*m*-OCH_3), 60.0 (CH_3, *p*-OCH_3), 94.8 (C, aromatic C), 101.8 (C, aromatic C), 104.5 (2CH, 2×aromatic CH), 109.8 (CH, aromatic CH), 119.1 (CH, aromatic CH), 119.4 (CH, aromatic CH), 121.4 (CH, aromatic CH), 127.3 (C, aromatic C), 127.8 (C, aromatic C), 130.7 (CH, aromatic CH), 136.8 (C, aromatic C), 137.8 (C, aromatic C), 139.9 (C, aromatic C), 144.3 (C, aromatic C), 148.5 (2C, 2×aromatic C), 152.4 (C, C=O), 153.5 (C, C=O); *m*/*z* (ES^+) 448.1 $[M+H]^+$, (40%); HRMS (ES^+) exact mass calculated for $C_{23}H_{22}N_5O_5$ $[M+H]^+$ 448.1621. Found 448.1611.

*4.1.16. N,N-Diethyl-1-(1H-pyrrolo[2,3-b]pyridin-3-yl)methanamine (**33**).* A solution of diethylamine hydrochloride (0.604 g, 5.5 mmol) and water (5 mL) was cooled in an ice bath to 0 °C. This solution was treated with 37 wt% aq formaldehyde (0.36 mL, 4.8 mmol) and stirred for 30 min. 7-Azaindole (0.500 g, 4.2 mmol) in ethanol (10 mL) was then added, with the resulting mixture being maintained at 0 °C for 30 min, before being heated to reflux for 20 h. Once cooled to room temperature, the reaction mixture was diluted with water (20 mL), before the pH of the solution was adjusted to 11 using 50 wt% aq NaOH. Extraction with dichloromethane (3×20 mL) was then performed, and the combined organic extracts were dried over magnesium sulfate, filtered and evaporated under reduced pressure to yield the crude product as a yellow/orange residue. The crude product was then subjected to flash column chromatography (ethyl acetate/methanol, 80:20) to give the pure product **33** as a reddish brown solid (0.755 g, 88%): mp 62–64 °C; R_f (10% methanol/ethyl acetate) 0.18; ν_{max}/cm^{-1} (KBr) 3142, 2970, 2931, 2787, 1583, 1544, 1454, 1421, 1298, 1048; δ_H (300 MHz, DMSO-d_6) 0.96–1.00 [6H, t, *J* 7.1, 2×NCH_2CH_3], 2.40–2.44 [4H, q, *J* 7.1, 2×NCH_2CH_3], 3.66 [2H, s, C–H_2], 6.99–7.03 [1H, q, *J* 7.8, 4.7, C–H_5], 7.32 [1H, s, C–H_2], 7.98–8.01 [1H, dd, *J* 7.8, 1.2, C–H_4], 8.17–8.19 [1H, dd, *J* 4.6, 1.4, C–H_6], 11.44 [1H, br s, NH]; δ_C (75 MHz, DMSO-d_6) 11.8 ($2CH_3$, 2×NCH_2CH_3), 45.9 ($2CH_2$, 2×NCH_2CH_3), 48.1 (CH_2, $CH_2N(C_2H_5)_2$), 110.9 (C, aromatic C), 114.8 (CH, aromatic CH), 119.7 (C, aromatic C), 124.5 (CH, aromatic CH), 127.3 (CH, aromatic CH), 142.3 (CH, aromatic CH), 148.8 (C, aromatic C); *m*/*z* (ES^+) 204.1 $[M+H]^+$ (100%); HRMS (ES^+) exact mass calculated for $C_{12}H_{18}N_3$ $[M+H]^+$, 204.1501. Found 204.1498.

*4.1.17. N,N-Dimethyl-1-(1H-pyrrolo[2,3-b]pyridin-3-yl)methanamine (**31**)[39].* A mixture of dimethylamine (33 wt% in EtOH, 9.8 mL, 55.2 mmol), acetic acid (2.7 mL, 48 mmol) and water (10 mL) was cooled in an ice bath to 0 °C. The resulting solution was subsequently treated with 37 wt% aq formaldehyde (3.6 mL, 48.1 mmol) and stirred for 30 min. 7-Azaindole (5.00 g, 42.4 mmol) in ethanol (20 mL) was then added, with the resulting solution being stirred at 0 °C for 30 min, before being heated to 100 °C for 18 h, at which point TLC analysis showed complete consumption of starting material. Once cooled to room temperature, the solution was diluted with water (40 mL), before its pH was adjusted to 11 using 50 wt% aq NaOH. After extraction with dichloromethane (3×30 mL), the organic extracts were dried over magnesium sulfate, filtered and evaporated under reduced pressure to give the crude product as a pale yellow solid. Purification was carried out by flash column chromatography (ethyl acetate/methanol, 80:20) to yield the pure product **31** as a white crystalline solid (6.05 g, 82%): mp 157–158 °C (lit.[39] 144–152 °C); R_f (20% methanol/ethyl acetate) 0.14; ν_{max}/cm^{-1} (KBr) 3079, 2947, 2775, 1604, 1528, 1412, 1334, 1002; δ_H (300 MHz, DMSO-d_6) 2.13 [6H, s, 2×NCH_3], 3.51 [2H, s, C–H_2], 7.00–7.04 [1H, q, *J* 7.9, 4.7, C–H_5], 7.33 [1H, s, C–H_2], 7.96–7.99 [1H, dd, *J* 7.9, 1.5, C–H_4], 8.18–8.20 [1H, dd, *J* 4.7, 1.5, C–H_6], 11.46 [1H, br s, NH]; δ_C (75 MHz, DMSO-d_6) 44.8 ($2CH_3$, 2×NCH_3), 54.4 (CH_2, $CH_2N(CH_3)_2$), 110.6 (C, aromatic C), 114.9 (C, aromatic CH), 119.7 (C, aromatic C), 124.7 (CH, aromatic CH), 127.2 (CH, aromatic CH), 142.4 (CH, aromatic CH), 148.7 (C, aromatic C); *m*/*z* (ES^+) 176.1 $[M+H]^+$ (100%).

*4.1.18. 2-(1H-Pyrrolo[2,3-b]pyridin-3-yl)acetonitrile (**32**)[40].* To a solution of *N*,*N*-dimethyl-1-(1*H*-pyrrolo[2,3-*b*]pyridin-3-yl)methanamine **30** (5.60 g, 32.0 mmol) in DMF (20 mL) was added a solution of sodium cyanide (2.35 g, 48.2 mmol) in water (16 mL). Acetic acid (5 mL) was then added to the mixture in a dropwise manner, and the resulting solution was heated to 110 °C for 8 h. After cooling to room temperature, the solution was diluted with saturated aq K_2CO_3 (30 mL) and extracted with ethyl acetate (3×40 mL). The organic extracts were dried over magnesium sulfate, filtered and evaporated under reduced pressure to yield the crude product. Purification by flash column chromatography (ethyl acetate/hexane 50:50) gave the pure product **32** as a white crystalline solid (2.59 g, 52%): mp 135–137 °C (lit.[40] 136–138 °C); R_f (50% ethyl acetate/hexane) 0.19; ν_{max}/cm^{-1} (KBr) 3089, 2888, 2248, 1611, 1585, 1538, 1420, 1337; δ_H (300 MHz, $CDCl_3$) 3.85 [2H, s, C–H_2], 7.14–7.18 [1H, q, *J* 7.9, 4.8, C–H_5], 7.40 [1H, s, C–H_2], 7.97–8.01 [1H, dd, *J* 7.9, 1.5, C–H_4], 8.37–8.39 [1H, dd, *J* 4.8, 1.5, C–H_6], 11.92 [1H, br s, NH]; δ_C (75 MHz, $CDCl_3$) 14.6 (CH_2, CH_2CN), 102.8 (C, aromatic C), 116.0 (CH, aromatic CH), 117.8 (C, CH_2CN), 119.0 (C, aromatic C), 124.0 (CH, aromatic CH), 127.1 (CH, aromatic CH), 143.1 (CH, aromatic CH), 148.9 (C, aromatic C); *m*/*z* (ES^+) 158.0 $[M+H]^+$ (100%).

*4.1.19. 2-(1-Methyl-1H-pyrrolo[2,3-b]pyridin-3-yl)acetonitrile (**34**)[41].* 2-(1*H*-Pyrrolo[2,3-*b*]pyridin-3-yl)acetonitrile **32** (2.11 g, 13.4 mmol) was dissolved in anhydrous DMF (30 mL) and the resulting solution was cooled to 0 °C in an ice bath. Sodium hydride (55 wt%, 0.585 g, 13.4 mmol) was added slowly in a portionwise manner. Once the addition was complete, the resulting mixture was allowed to stir at 0 °C for 20 min. At this point, a solution of iodomethane (0.83 mL, 13.4 mmol) in DMF (5 mL) was added in a dropwise fashion. The reaction mixture was then allowed to warm to room temperature, with stirring, over 2 h. Water (20 mL) was cautiously added to quench to reaction. Following extraction with ethyl acetate (3×30 mL), the combined organic extracts were washed with water (4×50 mL) and brine (50 mL) then dried over magnesium sulfate. The solvent was evaporated under reduced pressure to yield the crude product as a brown solid, which was then purified by flash column chromatography (hexane/ethyl acetate, 70:30) to give the pure product **34** as a reddish solid (1.06 g, 46%): mp 82–84 °C (lit.[41] 81–82.5 °C); R_f (30% ethyl acetate/hexane) 0.32; ν_{max}/cm^{-1} (NaCl) 2941, 2360, 2249, 1602, 1542, 1463, 1410, 1301, 1144; δ_H (400 MHz, $CDCl_3$) 3.83 [2H, s, C–H_2], 3.89 [3H, s, CH_3], 7.11–7.14 [1H, q, *J* 7.9, 4.7, C–H_5], 7.22 [1H, s, C–H_2], 7.90–7.93 [1H, dd, *J* 7.9, 1.5, C–H_4], 8.39–8.40 [1H, dd, *J* 4.7, 1.5, C–H_6]; δ_C (75 MHz, $CDCl_3$) 14.5 (CH_2, CH_2CN), 31.3 (CH_3, NCH_3), 101.5 (C, aromatic C), 115.8 (CH, aromatic CH), 117.8 (C, CH_2CN), 118.8 (C, aromatic C), 126.6 (CH, aromatic CH), 127.5 (CH, aromatic CH), 143.8 (CH, aromatic CH), 147.9 (C, aromatic C); *m*/*z* (ES^+) 172.1 $[M+H]^+$ (100%).

*4.1.20. 2-(1-Methyl-1H-pyrrolo[2,3-b]pyridin-3-yl)-3-oxo-3-(3,4,5-trimethoxyphenyl)propanenitrile (**35**).* A solution of LDA (1.8 M, 6.49 mL, 11.7 mmol) in THF (40 mL) was cooled to −78 °C under a nitrogen atmosphere, to which a solution of acetonitrile **34** (0.910 g, 5.3 mmol) in THF (5 mL) was added in a dropwise manner and the reaction mixture was stirred for 1 h, with the temperature maintained at −78 °C. A solution of 3,4,5-trimethoxybenzoyl chloride (1.48 g, 6.4 mmol) in THF (10 mL) was added dropwise to

the reaction mixture, which was then allowed to slowly warm to room temperature and was stirred for 12 h. The solvent was then removed under reduced pressure and the residue was dissolved in ethyl acetate (50 mL) and was subsequently washed with saturated aqueous sodium bicarbonate solution (40 mL), water (40 mL) and brine (40 mL). After being dried over magnesium sulfate, the solvent was removed under reduced pressure. The crude residue was subjected to flash column chromatography (hexane/ethyl acetate, 80:20), which yielded the pure product **35** as a yellow solid (0.956 g, 49%): mp 80–82 °C; R_f (80% ethyl acetate/hexane) 0.51; ν_{max}/cm^{-1} (KBr) 2940, 2201, 1684, 1583, 1505, 1461, 1415, 1333, 1127; δ_H (400 MHz, $CDCl_3$) 3.83 [6H, s, 2×*m*-OCH_3], 3.87 [3H, s, *p*-OCH_3], 3.89 [3H, s, NCH_3], 5.82 [1H, s, CH], 7.13–7.16 [1H, q, *J* 8.0, 4.7, C–H_5], 7.22 [2H, s, C–$H_{2',6'}$], 7.30 [1H, s, C–H_2], 8.04–8.06 [1H, dd, *J* 8.0, 1.5, C–H_4], 8.37–8.38 [1H, dd, *J* 4.7, 1.3, C–H_6]; δ_C (75 MHz, $CDCl_3$) 31.5 (CH_3, NCH_3), 38.6 (CH, *C*HCN), 56.4 ($2CH_3$, 2×*m*-OCH_3), 61.0 (CH_3, *p*-OCH_3), 102.4 (C, aromatic C), 105.6 (C, aromatic C), 106.8 (2CH, 2×aromatic CH), 116.5 (CH, aromatic CH), 118.0 (C, CN), 127.1 (CH, aromatic CH), 128.3 (CH, aromatic CH), 128.3 (C, aromatic C), 143.8 (C, aromatic C), 144.2 (CH, aromatic CH), 147.8 (C, aromatic C), 153.1 (2C, 2×aromatic C), 187.2 (C, C=O); *m*/*z* (ES^+) 366.1 $[M+H]^+$, (100%); HRMS (ES^+) exact mass calculated for $C_{20}H_{20}N_3O_4$ $[M+H]^+$ 366.1454. Found 366.1448.

*4.1.21. 5-Amino-4-(1-methyl-1H-pyrrolo[2,3-b]pyridin-3-yl)-3-(3,4,5-trimethoxyphenyl)-1H-pyrazole (**12**).* To a solution of ketonitrile **35** (0.806 g, 2.2 mmol) in absolute alcohol (30 mL), HCl (12 M, 1 mL) was slowly added. Hydrazine monohydrate (50%, 0.69 mL, 11.0 mmol) was then added and the resulting mixture was heated to reflux for 20 h. The reaction mixture was cooled to room temperature and the solvent was removed under reduced pressure. The residue was dissolved in ethyl acetate (40 mL) and was then washed with saturated aqueous sodium bicarbonate solution (30 mL), water (30 mL) and brine (30 mL), before being dried over magnesium sulfate. The solvent was evaporated under reduced pressure and the crude product was then purified by flash column chromatography (ethyl acetate/methanol, 90:10) to yield the pure product **12** as a brown solid (0.586 g, 70%): mp 266–267 °C; R_f (10% methanol/ethyl acetate) 0.40; ν_{max}/cm^{-1} (KBr) 3357, 3205, 2935, 1579, 1516, 1459, 1410, 1248, 1124; δ_H (400 MHz, DMSO-d_6) 3.37 [6H, s, 2×*m*-OCH_3], 3.59 [3H, s, *p*-OCH_3], 3.86 [3H, s, NCH_3], 4.51 [2H, br s, NH_2], 6.71 [2H, s, C–$H_{2',6'}$], 6.95–6.98 [1H, q, *J* 7.9, 4.7, C–H_5], 7.35–7.37 [1H, dd, *J* 7.8, 1.5, C–H_4], 7.56 [1H, s, C–H_2], 8.23–8.25 [1H, dd, *J* 4.6, 1.5, C–H_6], 11.97 [1H, br s, NH]; δ_C (75 MHz, DMSO-d_6) 30.8 (CH_3, NCH_3), 55.3 ($2CH_3$, 2×*m*-OCH_3), 60.1 (CH_3, *p*-OCH_3), 95.2 (C, broad aromatic C), 103.9 (2CH, 2×aromatic CH), 104.4 (C, aromatic C), 115.1 (CH, aromatic CH), 119.5 (C, aromatic C), 126.9 (C, broad aromatic C), 128.0 (CH, aromatic CH), 129.2 (CH, aromatic CH), 136.9 (C, aromatic C), 141.8 (C, broad aromatic C), 142.4 (CH, aromatic CH), 147.5 (C, aromatic C), 152.3 (C, broad aromatic C), 152.5 (2C, 2×aromatic C); *m*/*z* (ES^+) 380.1 $[M+H]^+$ (100%); HRMS (ES^+) exact mass calculated for $C_{20}H_{22}N_5O_3$ $[M+H]^+$ 380.1723. Found 380.1725.

*4.1.22. 5-Amino-N-methyl-4-(1-methyl-1H-pyrrolo[2,3-b]pyridin-3-yl)-3-(3,4,5-trimethoxyphenyl)-1H-pyrazole-1-carbothioamide (**36**).* To a solution of aminopyrazole **12** (0.080 g, 0.21 mmol) in acetonitrile (10 mL) was added methyl isothiocyanate (0.016 g, 0.22 mmol). The reaction mixture was then heated at reflux for 26 h before being cooled to room temperature. The solvent was removed under reduced pressure to give a crude brown residue. Purification by flash column chromatography (hexane/ethyl acetate, 50:50) yielded the pure product **36** as a white solid (0.044 g, 47%): mp 169–171 °C; R_f (50% ethyl acetate/hexane) 0.19; ν_{max}/cm^{-1} (KBr) 3320, 2933, 1626, 1594, 1523, 1469, 1423, 1361, 1284, 1123; δ_H (300 MHz, DMSO-d_6) 3.16 [3H, br d, *J* 3.8, NHC*H*$_3$], 3.37 [6H, s, 2×*m*-OCH_3], 3.59 [3H, s, *p*-OCH_3], 3.88 [3H, s, NCH_3], 6.90 [2H, s, C–$H_{2',6'}$], 7.01–7.05 [1H, q, *J* 7.8, 4.6, C–H_5], 7.14 [2H, br s, NH_2], 7.49–7.52 [1H, dd, *J* 7.8, 1.5, C–H_4], 7.60 [1H, s, C–H_2], 8.27–8.29 [1H, dd, *J* 4.6, 1.5, C–H_6], 10.15 [1H, br d, *J* 4.2, N*H*CH_3]; δ_C (75 MHz, DMSO-d_6) 30.8 (CH_3, NCH_3), 31.1 (CH_3, $NHCH_3$), 55.2 ($2CH_3$, 2×*m*-OCH_3), 60.0 (CH_3, *p*-OCH_3), 92.7 (C, aromatic C), 102.8 (C, aromatic C), 104.9 (2CH, 2×aromatic CH), 115.4 (CH, aromatic CH), 119.9 (C, aromatic C), 127.5 (C, aromatic C), 127.6 (CH, aromatic CH), 130.3 (CH, aromatic CH), 137.7 (C, aromatic C), 142.6 (CH, aromatic CH), 147.7 (C, aromatic C), 148.9 (C, aromatic C), 150.2 (C, aromatic C), 152.3 (2C, 2×aromatic C), 175.7 (C=S); *m*/*z* (ES^+) 453.1 $[M+H]^+$, (100%); HRMS (ES^+) exact mass calculated for $C_{22}H_{25}N_6O_3S$ $[M+H]^+$ 453.1709. Found 453.1700.

*4.1.23. 1-[5-Amino-4-(1-methyl-1H-pyrrolo[2,3-b]pyridin-3-yl)-3-(3,4,5-trimethoxyphenyl)-1H-pyrazol-1-yl]ethanone (**37**).* To a solution of aminopyrazole **12** (0.080 g, 0.21 mmol) in acetonitrile (10 mL) was added acetic anhydride (0.021 mL, 0.22 mmol). The reaction mixture was heated to reflux for 24 h before being cooled to room temperature. The solvent and excess acetic anhydride was then removed under reduced pressure. The crude residue was subjected to flash column chromatography (hexane/ethyl acetate, 40:60), yielding the pure product **37** as a pale yellow crystalline solid (0.055 g, 62%): mp 138–140 °C; R_f (60% ethyl acetate/hexane) 0.26; ν_{max}/cm^{-1} (KBr) 3435, 2939, 1702, 1636, 1597, 1577, 1470, 1421, 1380, 1351, 1128; δ_H (300 MHz, $CDCl_3$) 2.79 [3H, s, $COCH_3$], 3.48 [6H, s, 2×*m*-OCH_3], 3.79 [3H, s, *p*-OCH_3], 3.90 [3H, s, NCH_3], 5.60 [2H, br s, NH_2], 6.81 [2H, s, C–$H_{2',6'}$], 6.99–7.04 [1H, q, *J* 7.9, 4.7, C–H_5], 7.15 [1H, s, C–H_2], 7.60–7.63 [1H, dd, *J* 7.9, 1.5, C–H_4], 8.34–8.36 [1H, dd, *J* 4.7, 1.4, C–H_6]; δ_C (75 MHz, $CDCl_3$) 23.3 (CH_3, $COCH_3$), 31.3 (CH_3, NCH_3), 55.7 ($2CH_3$, 2×*m*-OCH_3), 60.9 (CH_3, *p*-OCH_3), 93.9 (C, aromatic C), 103.5 (C, aromatic C), 104.9 (2CH, 2×aromatic CH), 115.9 (CH, aromatic CH), 119.9 (C, aromatic C), 127.8 (C, aromatic C), 128.5 (2CH, 2×aromatic CH), 138.5 (C, aromatic C), 143.6 (CH, aromatic CH), 147.9 (C, aromatic C), 148.7 (C, aromatic C), 152.9 (2C, 2×aromatic C), 153.1 (C, aromatic C), 173.9 (C, C=O); *m*/*z* (ES^+) 422.2 $[M+H]^+$, (100%); HRMS (ES^+) exact mass calculated for $C_{22}H_{23}N_5O_4$ $[M+H]^+$ 422.1828. Found 422.1812.

*4.1.24. 2,2,2-Trifluoro-N-(4-(1-methyl-1H-pyrrolo[2,3-b]pyridin-3-yl)-3-(3,4,5-trimethoxyphenyl)-1H-pyrazol-5-yl)acetamide (**38**).* To a solution of aminopyrazole **12** (0.080 g, 0.21 mmol) in acetonitrile (10 mL) was added trifluoroacetic anhydride (0.04 mL, 0.25 mmol). The reaction mixture was then heated to reflux for 28 h before being left to cool to room temperature. Evaporation of the solvent under reduced pressure left the crude product as a brown residue. This residue was then purified by flash column chromatography (hexane/ethyl acetate, 40:60), which yielded the product **38** as a pale yellow solid (0.041 g, 41%): mp 230–231 °C; R_f (60% ethyl acetate/hexane) 0.25; ν_{max}/cm^{-1} (KBr) 3307, 2939, 1714, 1579, 1507, 1464, 1407, 1173, 1129; δ_H (300 MHz, DMSO-d_6) 3.49 [6H, s, 2×*m*-OCH_3], 3.69 [3H, s, *p*-OCH_3], 3.92 [3H, s, NCH_3], 6.82 [2H, s, C–$H_{2',6'}$], 7.03–7.07 [1H, q, *J* 7.9, 4.7, C–H_5], 7.42–7.45 [1H, dd, *J* 7.9, 1.5, C–H_4], 7.54 [1H, s, C–H_2], 8.31–8.34 [1H, dd, *J* 4.6, 1.4, C–H_6], 11.23 [1H, br s, $NHCOCF_3$], 13.52 [1H, br s, pyrazole NH]; δ_C (75 MHz, DMSO-d_6) 30.8 (CH_3, NCH_3), 55.4 ($2CH_3$, 2×*m*-OCH_3), 60.0 (CH_3, *p*-OCH_3), 102.8 (C, aromatic C), 104.0 (2C, 2×aromatic CH), 106.5 (C, aromatic C), 106.7 (C, aromatic C), 115.8 (C, q, J_{F-C} 288.4, CF_3), 115.3 (CH, aromatic CH), 118.7 (C, aromatic C), 124.5 (C, aromatic C), 127.8 (CH, aromatic CH), 128.8 (CH, aromatic CH), 137.3 (C, aromatic C), 142.6 (CH, aromatic CH), 147.4 (C, aromatic C), 152.7 (2C, 2×aromatic C), 153.1 (C, aromatic C), 156.3 (C, q, J_{F-C} 36.4, C=O); *m*/*z* (ES^+) 476.1 $[M+H]^+$, (100%); HRMS (ES^+) exact mass calculated for $C_{22}H_{21}N_5O_4F_3$ $[M+H]^+$ 476.1546. Found 476.1540.

4.1.25. 8-(1-Methyl-1H-pyrrolo[2,3-b]pyridin-3-yl)-7-(3,4,5-trimethoxyphenyl)pyrazolo[1,5-a][1,3,5]triazine-2,4(1H,3H)-dione

(***39***). Aminopyrazole **12** (0.080 g, 0.21 mmol) was dissolved in DCM (10 mL) containing two drops of triethylamine, and the resulting solution was cooled to 0 °C in an ice bath. This solution was then treated, while stirring, with *N*-chlorocarbonyl isocyanate (0.02 mL, 0.23 mmol). The reaction mixture was allowed to warm to room temperature and was stirred for 15 h. After careful addition of water (3 mL), the precipitate formed was filtered off before subsequently being washed with water (4×5 mL) and ether (2×5 mL). The precipitate was dessicated at 50 °C overnight to give the pure product **39** as a white solid (0.053 g, 56%): mp 301–303 °C; R_f (20% methanol/ethyl acetate) 0.62; ν_{max}/cm^{-1} (KBr) 3516, 2834, 1765, 1703, 1648, 1583, 1549, 1421, 1332, 1123; δ_H (400 MHz, DMSO-d_6) 3.37 [6H, s, 2×*m*-OCH_3], 3.61 [3H, s, *p*-OCH_3], 3.88 [3H, s, NCH_3], 6.84 [2H, s, C–$H_{2',6'}$], 7.02–7.06 [1H, q, *J* 7.8, 4.6, C–H_5], 7.50 [1H, d, *J* 7.8, C–H_4], 7.64 [1H, s, C–H_2], 8.29 [d, 1H, *J* 4.5, C–H_6], 11.66 [2H, br s, 2×NH]; δ_C (75 MHz, DMSO-d_6) 30.9 (CH_3, NCH_3), 55.2 ($2CH_3$, 2×*m*-OCH_3), 60.0 (CH_3, *p*-OCH_3), 94.2 (C, aromatic C), 100.4 (C, aromatic C), 104.5 (2CH, 2×aromatic CH), 115.6 (CH, aromatic CH), 119.9 (C, aromatic C), 127.2 (C, aromatic C), 127.6 (C, aromatic CH), 131.0 (CH, aromatic CH), 137.9 (C, aromatic C), 140.1 (C, aromatic C), 142.7 (CH, aromatic CH), 144.3 (C, aromatic C), 147.6 (C, aromatic C), 148.6 (C, C=O), 152.5 (2C, 2×aromatic C), 153.4 (C, C=O); *m*/*z* (ES^-) 447.1 $[M-H]^-$, (100%); HRMS (ES^+) exact mass calculated for $C_{22}H_{21}N_6O_5$ $[M+H]^+$ 449.1573. Found 449.1554.

*4.1.26. 3-(1-Methyl-1H-pyrrolo[2,3-b]pyridin-3-yl)-5,7-bis(trifluoromethyl)-2-(3,4,5-trimethoxyphenyl)pyrazolo[1,5-a]pyrimidine (**40**).* To a stirred mixture of aminopyrazole **12** (0.080 g, 0.21 mmol) in acetic acid (10 mL) was added hexafluoroacetylacetone (0.60 mL, 4.20 mmol). This mixture was then heated at 80 °C for 20 h. After cooling to room temperature, the acetic acid was removed under reduced pressure, and the residue was dissolved in ethyl acetate (15 mL) and then washed with water (10 mL) and brine (10 mL) and dried over magnesium sulfate. Evaporation of the solvent under reduced pressure yielded the crude product as a red/brown solid. Purification via flash column chromatography (hexane/ethyl acetate, 50:50) gave the pure product **40** as an orange solid (0.062 g, 54%): mp 174–176 °C; R_f (50% ethyl acetate/hexane) 0.46; ν_{max}/cm^{-1} (KBr) 2926, 1590, 1481, 1419, 1271, 1127, 1010; δ_H (400 MHz, $CDCl_3$) 3.55 [6H, s, 2×*m*-OCH_3], 3.86 [3H, s, *p*-OCH_3], 4.02 [3H, s, NCH_3], 6.94–6.97 [1H, q, *J* 7.6, 4.6, C–H_5], 7.04 [2H, s, C–$H_{2',6'}$], 7.38 [1H, d, *J* 7.8, C–H_4], 7.46 [1H, s, (CF_3)C–CH=C(CF_3)], 7.56 [1H, s, C–H_2], 8.35 [1H, br s, C–H_4]; δ_C (75 MHz, $CDCl_3$) 31.6 (CH_3, NCH_3), 55.9 ($2CH_3$, 2×*m*-OCH_3), 60.9 (CH_3, *p*-OCH_3), 102.0 (C, aromatic C), 102.3 (CH, aromatic CH), 106.2 (C, aromatic C), 106.3 (2CH, 2×aromatic CH), 115.9 (CH, aromatic CH), 119.1 (C, q, J_{F-C} 275.4, CF_3), 120.3 (C, q, J_{F-C} 275.2, CF_3), 119.2 (C, aromatic C), 127.1 (C, aromatic C), 129.3 (CH, aromatic CH), 129.4 (CH, aromatic CH), 135.1 (C, q, J_{F-C} 34.7, aromatic C), 139.3 (C, aromatic C), 143.4 (CH, aromatic CH), 145.0 (C, q, J_{F-C} 37.8, aromatic C), 146.7 (C, aromatic C), 148.0 (C, aromatic C), 153.3 (2C, 2×aromatic C), 156.0 (C, aromatic C); *m*/*z* (ES^+) 552.1 $[M+H]^+$, (100%); HRMS (ES^+) exact mass calculated for $C_{25}H_{20}N_5O_3F_6$ $[M+H]^+$ 552.1470. Found 552.1469.

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References and notes

1. Hopkins, A. L.; Groom, C. R. *Nat. Rev. Drug Discovery* **2002**, *1*, 727–730.
2. Daub, H. *Biochim. Biophys. Acta, Proteins Proteomics* **2005**, *1754*, 183–190.
3. Kornev, A. P.; Haste, N. M.; Taylor, S. S.; Eyck, L. F. T. *Proc. Natl. Acad. Sci. U.S.A.* **2006**, *103*, 17783–17788.
4. (a) Sanchez, C.; Mendez, C.; Salas, J. A. *Nat. Prod. Rep.* **2006**, *23*, 1007–1045; (b) Lapenna, S.; Giordano, A. *Nat. Rev. Drug Discovery* **2009**, *8*, 547–566.
5. (a) Marengo, B.; De, C.; Ricciarelli, R.; Pronzato, M.; Marinari, U. M.; Domenicotti, C. *Cancer.* **2011**, *3*, 531–567; (b) Gani, O.; Engh, R. *Nat. Prod. Rep.* **2010**, *27*, 489–498.
6. Peifer, C.; Wagner, G.; Laufer, S. *Curr. Top. Med. Chem.* **2006**, *6*, 113–149.
7. Schwid, S.; Shoulson, I.; Marek, K.; Oakes, D.; Kieburtz, K.; Gorbold, E.; Fahn, S.; Goetz, C.; Rudolph, A.; Shinaman, A. *Neurology* **2004**, *62*, 330–332.
8. Grimminger, F.; Schermuly, R. T.; Ghofrani, H. A. *Nat. Rev. Drug Discovery* **2010**, *9*, 956–970.
9. (a) Janne, P. A.; Gray, N.; Settleman, J. *Nat. Rev. Drug Discovery* **2009**, *8*, 709–723; (b) Sawa, M. *Mini-Rev. Med. Chem.* **2008**, *8*, 1291–1297.
10. Aiello, L. P.; Clermont, A.; Arora, V.; Davis, M. D.; Sheetz, M. J.; Bursell, S. E. *Invest. Ophthalmol. Visual Sci.* **2006**, *47*, 86–92.
11. Grant, S.; Tran, P.; Zhang, Q.; Zou, A.; Dinh, D.; Jensen, J.; Zhou, S.; Kang, X.; Zachwieja, J.; Lippincott, J.; Liu, K.; Johnson, S. L.; Scales, S.; Yin, C.; Nukui, S.; Stoner, C.; Prasanna, G.; Lafontaine, J.; Wells, P.; Li, H. *Eur. J. Pharmacol.* **2010**, *627*, 16–25.
12. Nettleton, D. E.; Doyle, T. W.; Krishnan, B.; Matsumoto, G. K.; Clardy, J. *Tetrahedron Lett.* **1985**, *26*, 4011–4014.
13. Tamaoki, T.; Nomoto, H.; Takahashi, I.; Kato, Y.; Morimoto, M.; Tomita, F. *Biochem. Biophys. Res. Commun.* **1986**, *135*, 397–402.
14. Gray, N.; Detivaud, L.; Doering, C.; Meijer, L. *Curr. Med. Chem.* **1999**, *6*, 859–875.
15. Peifer, C.; Stoiber, T.; Unger, E.; Totzke, F.; Schachtele, C.; Marmé, D.; Brenk, R.; Klebe, G.; Schollmeyer, D.; Dannhardt, G. *J. Med. Chem.* **2006**, *49*, 1271–1281.
16. Gassel, M.; Breitenlechner, C. B.; Konig, N.; Huber, R.; Engh, R. A.; Bossemeyer, D. *J. Biol. Chem.* **2004**, *279*, 23679–23690.
17. Davis, P. D.; Hill, C. H.; Lawton, G.; Nixon, J. S.; Wilkinson, S. E.; Hurst, S. A.; Keech, E.; Turner, S. E. *J. Med. Chem.* **1992**, *35*, 177–184.
18. Gaisina, I. N.; Gallier, F.; Ougolkov, A. V.; Kim, K. H.; Kurome, T.; Guo, S.; Holzle, D.; Luchini, D. N.; Blond, S. Y.; Billadeau, D. D.; Kozikowski, A. P. *J. Med. Chem.* **2009**, *52*, 1853–1863.
19. Pierce, L. T.; Cahill, M. M.; McCarthy, F. O. *Tetrahedron* **2010**, *66*, 9754–9761.
20. Carmeliet, P.; Jain, R. K. *Nature* **2000**, *407*, 249–257.
21. Peifer, C.; Selig, R.; Kinkel, K.; Ott, D.; Totzke, F.; Schachtele, C.; Heidenreich, R.; Rocken, M.; Schollmeyer, D.; Laufer, S. *J. Med. Chem.* **2008**, *51*, 3814–3824.
22. Peifer, C.; Krasowski, A.; Hammerle, N.; Kohlbacher, O.; Dannhardt, G.; Totzke, F.; Schachtele, C.; Laufer, S. *J. Med. Chem.* **2006**, *49*, 7549–7553.
23. Bossi, R. T.; Saccardo, M. B.; Ardini, E.; Menichincheri, M.; Rusconi, L.; Magnaghi, P.; Orsini, P.; Avanzi, N.; Borgia, A. L.; Nesi, M.; Bandiera, T.; Fogliatto, G.; Bertrand, J. A. *Biochemistry* **2010**, *49*, 6813–6825.
24. O'Neill, D. J.; Shen, L.; Prouty, C.; Conway, B. R.; Westover, L.; Xu, J. Z.; Zhang, H.-C.; Maryanoff, B. E.; Murray, W. V.; Demarest, K. T.; Kuo, G.-H. *Bioorg. Med. Chem.* **2004**, *12*, 3167–3185.
25. Zhang, H.-C.; Ye, H.; Conway, B. C.; Derian, C. K.; Addo, M. F.; Kuo, G. H.; Hecker, L. R.; Croll, J. L.; Westover, L.; Xu, J. Z.; Look, R.; Demarest, K. T.; Andrade-Gordon, P.; Damiano, B. P.; Maryanoff, B. E. *Bioorg. Med. Chem. Lett.* **2004**, *14*, 3245–3250.
26. Messaoudi, S.; Anizon, F.; Léonce, S.; Pierré, A.; Pfeiffer, B.; Prudhomme, M. *Eur. J. Med. Chem.* **2005**, *40*, 961–971.
27. Braña, M. F.; Cacho, M.; García, M. L.; Mayoral, E. P.; López, B.; de Pascual-Teresa, B.; Ramos, A.; Acero, N.; Llinares, F.; Muñoz-Mingarro, D.; Lozach, O.; Meijer, L. *J. Med. Chem.* **2005**, *48*, 6843–6854.
28. Pask, C. M.; Camm, K. D.; Kilner, C. A.; Halcrow, M. A. *Tetrahedron Lett.* **2006**, *47*, 2531–2534.
29. Liu, T.; Dong, X.; Xue, N.; Wu, R.; He, Q.; Yang, B.; Hu, Y. *Bioorg. Med. Chem.* **2009**, *17*, 6279–6285.
30. Faul, M. M.; Winneroski, L. L.; Krumrich, C. A. *J. Org. Chem.* **1998**, *63*, 6053–6058.
31. Braña, M. F.; Gradillas, A.; Ovalles, A. G.; Lopez, B.; Acero, N.; Llinares, F.; Mingarro, D. M. *Bioorg. Med. Chem.* **2006**, *14*, 9–16.
32. Exposito, A.; Fernandez-Suarez, M.; Iglesias, T.; Muñoz, L.; Riguera, R. *J. Org. Chem.* **2001**, *66*, 4206–4213.
33. Elgemeie, G. H.; El-Ezbawy, S. R.; Ali, H. A. *Synth. Commun.* **2001**, *31*, 3459–3467.
34. Anwar, H. F.; Elnagdi, M. H. *ARKIVOC* **2009**, *i*, 198–250.
35. Dai, H.-G.; Li, J.-T.; Li, T.-S. *Synth. Commun.* **2006**, *36*, 1829–1835.
36. Ahmad, A.; Eelnurme, I.; Spenser, I. D. *Can. J. Chem.* **1960**, *38*, 2523.
37. Bellemin, R.; Decerprit, J.; Festal, D. *Eur. J. Med. Chem.* **1996**, *31*, 123–132.
38. Pons, J.-F.; Fauchère, J.-L.; Lamaty, F.; Molla, A.; Lazaro, R. *Eur. J. Org. Chem.* **1988**, *5*, 853–860.
39. Robison, M. M.; Robison, B. L. *J. Am. Chem. Soc.* **1955**, *77*, 457–460.
40. Robison, M. M.; Robison, B. L. *J. Am. Chem. Soc.* **1956**, *78*, 1247–1251.
41. Bell, M. R. U.S. Patent 3,354,174, 1967.