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Structure-based design, synthesis and evaluation in vitro of arylnaphthyridinones, arylpyridopyrimidinones and their tetrahydro derivatives as inhibitors of the tankyrases

Katerina Kumpan [a], Amit Nathubhai [a], Chenlu Zhang [a,b], Pauline J. Wood [a], Matthew D. Lloyd [a], Andrew S. Thompson [a], Teemu Haikarainen [c], Lari Lehtiö [c], Michael D. Threadgill [a,*]

[a] Medicinal Chemistry, Department of Pharmacy & Pharmacology, University of Bath, Claverton Down, Bath BA2 7AY, UK
[b] Department of Pharmacy, Shandong University, Jinan 250100, China
[c] Biocenter Oulu and Faculty of Biochemistry and Molecular Medicine, University of Oulu, Oulu, Finland

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ABSTRACT

The tankyrases are members of the PARP superfamily; they poly(ADP-ribosyl)ate their target proteins using NAD^+ as a source of electrophilic ADP-ribosyl units. The three principal protein substrates of the tankyrases (TRF1, NuMA and axin) are involved in replication of cancer cells; thus inhibitors of the tankyrases may have anticancer activity. Using structure-based drug design and by analogy with known 3-arylisoquinolin-1-one and 2-arylquinazolin-4-one inhibitors, series of arylnaphthyridinones, arylpyridinopyrimidinones and their tetrahydro-derivatives were synthesised and evaluated in vitro. 7-Aryl-1,6-naphthyridin-5-ones, 3-aryl-2,6-naphthyridin-1-ones and 3-aryl-2,7-naphthyridin-1-ones were prepared by acid-catalysed cyclisation of the corresponding arylethynylpyridinenitriles or reaction of bromopyridinecarboxylic acids with β-diketones, followed by treatment with NH_3. The 7-aryl-1,6-naphthyridin-5-ones were methylated at 1-N and reduced to 7-aryl-1-methyl-1,2,3,4-tetrahydro-1,6-naphthyridin-5-ones. Cu-catalysed reaction of benzamidines with bromopyridinecarboxylic acids furnished 2-arylpyrido[2,3-*d*]pyrimidin-4-ones. Condensation of benzamidines with methyl 1-benzyl-4-oxopiperidine-3-carboxylate and deprotection gave 2-aryl-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidin-4-ones, aza analogues of the known inhibitor XAV939. Introduction of the ring-N in the arylnaphthyridinones and the arylpyridopyrimidinones caused >1000-fold loss in activity, compared with their carbocyclic isoquinolinone and quinazolinone analogues. However, the 7-aryl-1-methyl-1,2,3,4-tetrahydro-1,6-naphthyridin-5-ones showed excellent inhibition of the tankyrases, with some examples having $IC_{50} = 2$ nM. One compound (7-(4-bromophenyl)-1-methyl-1,2,3,4-tetrahydro-1,6-naphthyridin-5-one) showed 70-fold selectivity for inhibition of tankyrase-2 versus tankyrase-1. The mode of binding was explored through crystal structures of inhibitors in complex with tankyrase-2.

1. Introduction

The tankyrases (PARP-5a/TNKS-1/ARTD5 and PARP-5b/TNKS-2/ARTD6) are members of the poly(ADP-ribose)polymerase (PARP) superfamily of enzymes.[1] Considerable recent interest has been expressed in these enzymes,[2–10] in view of their roles at the chromosomal telomeres, at the mitotic spindle and in signalling pathways. In common with other PARPs, the tankyrases transfer ADP-ribose units from substrate NAD^+ to build poly(ADP-ribose)polymers on target proteins. TNKS-1 poly(ADP-ribosyl)ates TRF1 at the telomere, causing it to detach from the telomere and become ubiquitinylated and targeted for destruction.[11,12] This exposes the telomeric DNA, allowing access by the telomerase complex. Thus TNKS-1 activity is a positive regulator of telomere length. Tankyrase activity is also critical for the correct functioning of the mitotic spindle, where its protein target is NuMA.[13] TNKS-1 is also a component of the *wnt* signalling system,[9,14] where it modifies axin. This leads to ubiquitinylation and destruction of the poly(ADPr)-axin, increasing levels of β-catenin and suppressing phosphorylated β-catenin. The net effect of this activity is a proliferative signal in the nucleus. The isoform TNKS-2 can substitute for TNKS-1 in many of these intracellular activities.[11,15,16]

Tankyrases are over-expressed in several clinical cancers and cancer cell lines, including mammary[17] and colorectal carcinomas,[18] chronic myeloid leukaemia,[19] and tumours in brain,[20] stomach[21] and bladder.[22] This over-expression indicates critical roles for the tankyrases in tumours; they were proposed as therapeutic

* Corresponding author. Tel.: +44 1225 386840; fax: +44 1225 386114.
E-mail address: m.d.threadgill@bath.ac.uk (M.D. Threadgill).

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targets as long ago as 2006.[23] Their recent discovery[9] as components of the proliferative *wnt* signalling pathway confirms this potential. Indeed, one tankyrase inhibitor has shown activity in an APC-mutant mouse model of colon cancer[24] and has been shown to cause apoptosis in neuroblastoma cells.[8] The time is, therefore, ripe to explore the structure-based design and development of potent and selective inhibitors of the tankyrases.

The reported inhibitors of the tankyrases fall into two classes: those that bind in the nicotinamide-binding site as mimics of nicotinamide and those which are bound at other sites, including the adenine-binding site. Some recent inhibitors have been designed to occupy both sites simultaneously.

Examination of the crystal structure of the prototype inhibitor XAV939 **1** (Fig. 1) bound into TNKS-2 reveals that the lactam makes H-bonds with the carbonyl and N–H of Gly^{1032} and the side-chain alcohol of Ser^{1068}.[25] There is also some evidence for π-stacking with Tyr^{1071}. The side-chain 4-trifluoromethylphenyl projects into an adjacent hydrophobic cavity, forming a π-stack with Tyr^{1050} and hydrophobic interactions with Pro^{1034}, Phe^{1035} and Ile^{1075}.[4] We have reported that 2-aryl-8-methylquinazolin-4-ones **2** and related 3,4-dihydroquinazolinones also inhibit the tankyrases strongly, with IC_{50} values slightly higher than those of **1**.[4,26] Molecular modelling studies indicated that the mode of binding of **2** is similar to that of **1**, with the aryl group projecting into the hydrophobic cavity.[4] Very recently, we disclosed a large series of 5-substituted 3-arylisoquinolin-1-ones **3**, some of which (e.g., **3**: $R^{4'} = R^5 = Me$) were more potent than **1**. Water-soluble tertiary amine analogues (e.g., **3**: $R^{4'} = CH_2NMe_2$, $R^5 = Me$) retained potency.[3] A crystal structure of **3** ($R^{4'} = Cl$, $R^5 = OMe$) in complex with TNKS-2 (PDB code 4UVY) confirmed the mode of binding, with the lactam carbonyl oxygen accepting H-bonds from Gly^{1032} and Ser^{1068} and the lactam N–H donating an H-bond to the carbonyl of Gly^{1032}.[3] The chlorophenyl unit occupies the hydrophobic cavity, with the chlorine located at the entrance to a narrow tunnel leading to the exterior. Extensions at $R^{4'}$ project deep into this tunnel. Although the lactam carbonyl and the phenyl in the hydrophobic pocket are essential for binding, the N–H of the lactam is less important, in that flavones such as **4** are active.[27,28] The [1,2,4]triazolo[4,3-*b*]pyridazine **5** has also been shown crystallographically to bind in the nicotinamide-binding pocket, with 1-N making a H-bond to N–H of Gly^{1032}, 2-N making a H-bond with OH of Ser^{1068} and the exocyclic N–H bonding with the C=O of Gly^{1032}; the phenol occupies the hydrophobic cavity.[2] Curiously, this compound is selective for inhibition of TNKS-1 (IC_{50} = 12 nM) versus TNKS-2 (IC_{50} = 200 nM). IWR1 **6** binds solely at the nearby adenosine-binding site,[29] whereas **7**[7] and **8**[10] occupy both the nicotinamide-bind site and the adenosine-binding site.

2. Results and discussion

2.1. Design of inhibitors

In the light of the potency and selectivity of our series of 3-arylisoquinolin-1-ones **3**[3] and 2-arylquinazolin-4-ones **2**[4] for inhibition of the tankyrases, we sought to extend the structure–activity relationships (SARs) into aza analogues. Figure 2 shows the structures of these candidate inhibitors. Introducing a nitrogen into the carbocyclic ring of **3** (in naphthyridinones **9**–**11**) will have two potential effects, electron-deficiency in the bicycle and weak basicity or H-bonding involving the N-lone pair. *Para*-substitution was found to be optimum in the phenyl rings of **2**[4] and **3**[3]; thus only *para*-substituted phenyl rings were targeted in the present work. The lactam of **9**–**11** was expected to form the usual H-bonds with Ser^{1068} and Gly^{1032} (TNKS-2 numbering), mimicking the nicotinamide of NAD^+. The phenyl ring should occupy the adjacent hydrophobic cavity, making hydrophobic and π-stacking interactions therein, with the *para*-substituent projecting into the distal tunnel (Fig. 3 shows a cartoon of the binding pocket). In the 2-arylquinazolin-4-one series **2**, a small substituent, for example, Me, at the 8-position slightly improved potency.[3] Similarly, 5-methyl-3-arylisoquinolin-1-ones (**3**: $R^5 = Me$) generally had higher activity than their 5-fluoro (**3**: $R^5 = F$), 5-hydroxy (**3**: $R^5 = OH$) or 5-amino- (**3**: $R^5 = NH_2$) counterparts.[4] Thus we designed the 1-methylnaphthyridin-5-ones **12** with the *N*-methyl occupying the same pocket in the enzyme. Of course, this makes major changes in electron-distribution in the bicyclic core and **12** are cationic. Similarly, the N-oxide in **13** would occupy the same pocket but without overall charge. The arylpyridopyrimidinones **14**–**16** are aza analogues of the active quinazolinones **3**. As for **9**–**11**, the additional nitrogen may make H-bonds.

Unlike PARP-1, the nicotinamide-binding site of the tankyrases can accommodate a (partly) saturated ring mimicking the pyridinium of NAD^+, as shown by the activity of **1**. Thus we chose to explore the tetrahydro analogues **17** and **18**. In the 7-aryl-1-methyl-1,2,3,4-tetrahydro-1,6-naphthyridin-5-ones **17**, the left ring has three contiguous sp^2-hybridised atoms (presumably coplanar with the pyridinone) and three sp^3 methylenes. Thus the conformation may be slightly different to that of **1** but the N–Me should occupy the same space as the 5-Me in **3** ($R^5 = Me$) and the 8-Me in **2**. The tetrahydropyridopyrimidinones **18** (especially **18d**, $R = CF_3$) are strict analogues of **1**, where the neutral lipophilic sulfur is replaced by a secondary amine. This amine should be protonated at physiological pH, so **18** should have the same conformation as **1** but will test the tolerance of the binding pocket to positive charge.

2.2. Chemical synthesis

The initial targets for synthesis were the 7-aryl-1,6-naphthyridin-5-ones **9**, the 3-aryl-2,7-naphthyridin-1-ones **10** and the 3-aryl-2,6-naphthyridin-1-ones **11**. Wibberley prepared **9a** in 1.6% yield from ethyl 2-methylpyridine-3-carboxylate and benzaldehyde, via **26a**.[30] Reaction of 2-(2-phenyl-2-oxoethyl)pyridine-3-carboxylic acid with ammonia gives **9a** directly in 80% yield.[31] Nishiwaki et al. examined cyclisation of 2-phenylethynyl-3-cyanopyridine **25a** under various conditions.[32] Under basic conditions, the principal product arose from hydration/5-*exo*-dig cyclisation, whereas strong acid furnished an equimolar mixture of **26a** and **9a** from 6-*endo*-dig cyclisation of the

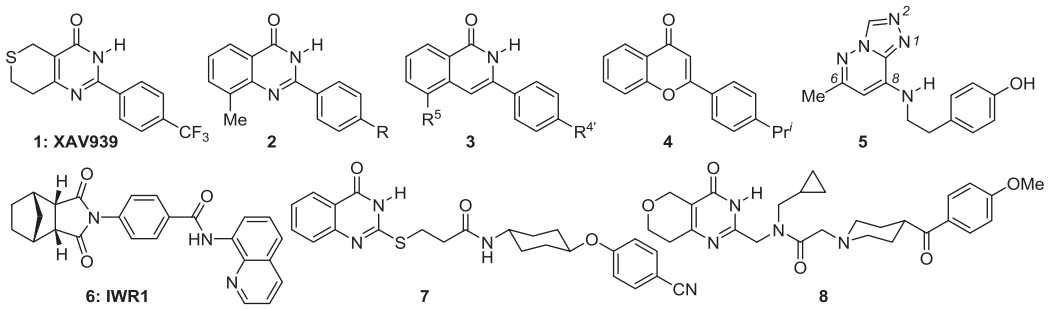


Figure 1. Structures of reported inhibitors of the tankyrases.

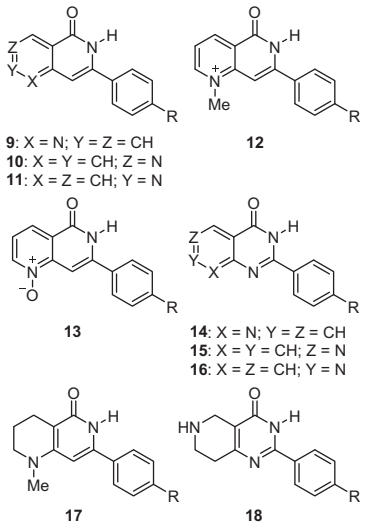


Figure 2. Structures of arylnaphthyridinones **9**–**13**, arylpyridopyrimidinones **14**–**16**, aryltetrahydronaphthyridinones **17** and aryltetrahydropyridopyrimidinones **18**, designed as inhibitors of the tankyrases. R = H, Me, OMe, CF_3, Cl, Br, NH_2, OH, —C≡CPh, *etc.*

intermediate amide. By contrast, Pt-catalysed cyclisation of **25a** is reported to afford a mixture of **9a** and 5-ethoxy-7-phenyl-1,6-naphthyridine in moderate yields.[33] This method also provides **10a** in low yield. Isomer **11a** has been prepared in low yield by debenzylation of 1-benzyl-4-phenacylpyridine-3-carboxamide.[35] This route was developed by N-benzylation of nicotinamide. Riessert attack of acetophenone enolate at the 4-position, acid-catalysed cyclisation and debenzylation yields **11a**.[35] 3-Phenyl-2,6-naphthyridin-1-one **10a** was formed in poor yield by a curious route involving treatment of dimethyl 2-(4-methoxycarbonylpyridin-3-yl)malonate with NaCl in DMSO at high temperature, followed by reaction with ammonia.[36]

Of these routes, we selected the acid-catalysed cyclisation of arylethynyl-cyanopyridines **25**, **28**, **35** (Scheme 1), as the diversity in the aryl group would be introduced through diversity in the starting arylethynes, many of which are commercially available or readily accessible synthetically. Arylethynes **21c**–**e**,**g**,**h**, which are not economically commercially available, were obtained by Sonogashira coupling of the corresponding iodoarenes **19c**–**e**,**g**,**h** with trimethylsilylethyne to give **20c**–**e**,**g**,**h**, followed by desilylation. The phenolic OH of 4-iodophenol had earlier been masked

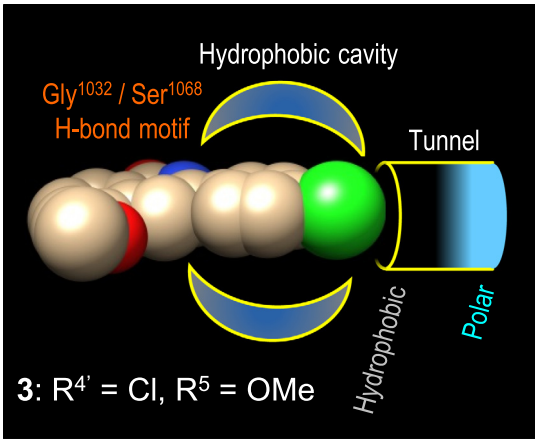
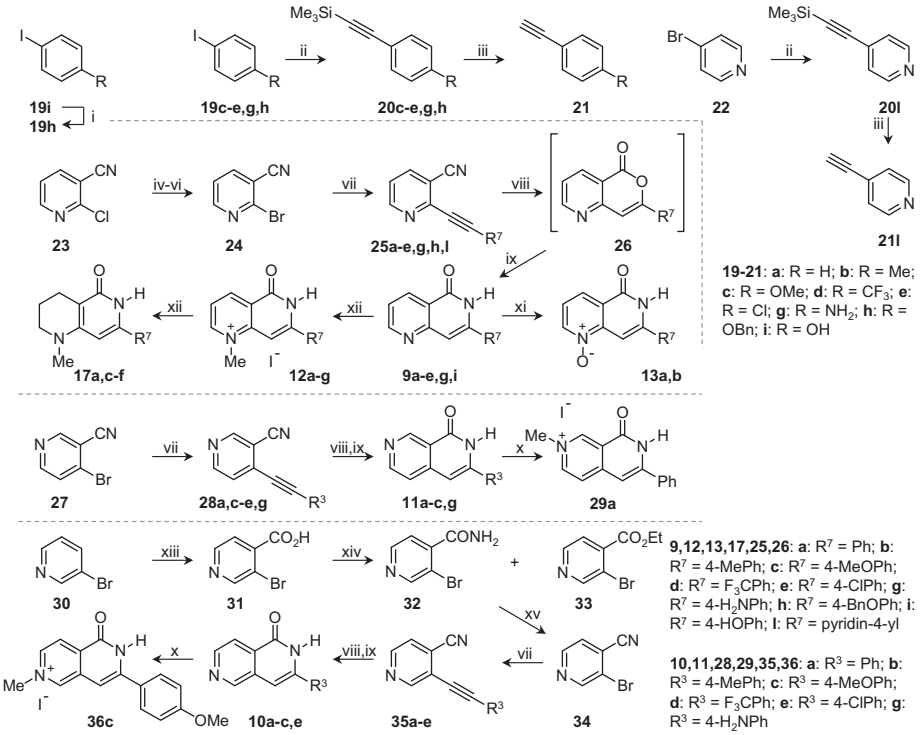


Figure 3. Cartoon showing the relative positions of binding motifs in the structures of the tankyrases, with the inhibitor **3** ($R^{4'}$ = Cl, R^5 = OMe) bound.

with benzyl, using benzyl bromide and caesium carbonate in DMF, giving **19h**. Similar coupling of 4-bromopyridine **22** with trimethylsilylethyne and subsequent desilylation afforded 4-ethynylpyridine **21l** in moderate yield, via intermediate **20l**. Sonogashira coupling of the alkynes **21a**–**e**,**g**,**h**,**l** with appropriate halopyridinenitriles was then investigated. Coupling of the alkynes with 2-chloro-3-cyanopyridine **23** led to the key intermediates **25** in low yields, despite activation of the chloroarene by the adjacent ring-N and the electron-withdrawing nitrile. This parallels our experience with Suzuki couplings to 3-chloroisoquinolines.[3] The poor yield was addressed in two ways. Firstly, 1,4-diarylbutadiynes were observed as major by-products, arising from oxidative Glaser homocoupling of the arylethynes.[37,38] This was addressed by using an excess of the alkyne (wasteful) or by adding sodium ascorbate to the reaction mixture as an antioxidant. Addition of sodium ascorbate also led to higher yields in some of the couplings of **19** and **22** with trimethylsilylethyne. Also effective in raising the yields of **25** was replacement of the 2-chloropyridine **23** with the corresponding bromopyridine **24**. This educt was prepared by acetolysis of **23** with acetic acid and hydrolysis of the intermediate 2-acetoxy-3-cyanopyridine in boiling aqueous THF under neutral conditions to give 3-cyanopyridin-2-one. Conventional reagents (e.g., $POBr_3$, PBr_5) failed to give **24** but reaction with Bu_4NBr and P_2O_5 in toluene gave the bromopyridine in excellent overall yield. This 2-bromopyridine coupled with the range of arylethynes in moderate-to-good yields to give **25a**–**e**,**g**,**h**,**l**, catalysed by $(Ph_3P)_2PdCl_2$. Aqueous acid-catalysed cyclisation proceeded exclusively in the 6-*endo*-dig mode, affording mixtures of the 7-arylpyrano[4,3-*b*]pyridin-5-ones **26** and the 7-aryl-1,6-naphthyridin-5-ones **9**, from initial hydration to the carboxamide and trapping of the electrophile derived from the alkyne with either the oxygen or the nitrogen, respectively, of the amide. In many cases, these mixtures were separable with difficulty and **26** could be converted into **9** by treatment with ammonia in 2-methoxyethanol under pressure. However, it was more convenient to treat the mixtures directly with ammonia to afford good yields of **9**. The benzyl protection of **25h** was lost during the treatment with acid, forming **9i** directly.

Methylation of **9a**–**e**,**g** at 1-N was achieved with iodomethane in DMF, affording **12a**–**e**,**g**. The location of the methyl at 1-N was confirmed by HMBC correlations from NCH_3 to 2-C and from 2-H to NCH_3 and, in some cases, by NOE experiments. Interestingly, **9g**, carrying a primary aniline, was only methylated at 1-N to give **12g**. These quaternised naphthyridinium salts **12** were targets in their own right but were also precursors of the tetrahydro analogues **17**. Borane·pyridine complex has been reported[39] to reduce naphthyridinium species but, in the case of **12**, it was necessary to use formic acid as solvent for the reaction. The problem of decomposition of the borane complex by the acidic medium was overcome by adding the reductant in frequent small portions and **17a**,**c**–**f** were obtained in low yields.

The nucleophilicity of 1-N was also exploited in forming the N-oxides **13a**,**b** from **9a**,**b**, using anhydrous peroxytrifluoroacetic acid generated in situ from urea·hydrogen peroxide complex and trifluoroacetic anhydride. We had previously used this system for the difficult N-oxidation of 8-cyanoquinoline to the N-oxide.[40] Although it was predictable that the oxidation had taken place at 1-N, in the light of methylation occurring exclusively at this site, it was important to provide evidence for the regiochemical identity of target compounds **13a**,**b**. In this case, the oxygen was located by examination of the ^{15}N NMR chemical shifts. After assignment using ^{1}H–^{15}N HMBC spectroscopy, the ^{15}N chemical shifts for **9a** were shown to be 1-N (δ 278.87, HMBC correlation to 2-H and 8-H) and 6-N (δ 124.89, HMBC correlation to 8-H only), whereas those for **13a** were 1-N (δ 252.76; HMBC correlation to 2-H and 8-H) and 6-N (δ 129.56; correlation to 8-H only). The marked



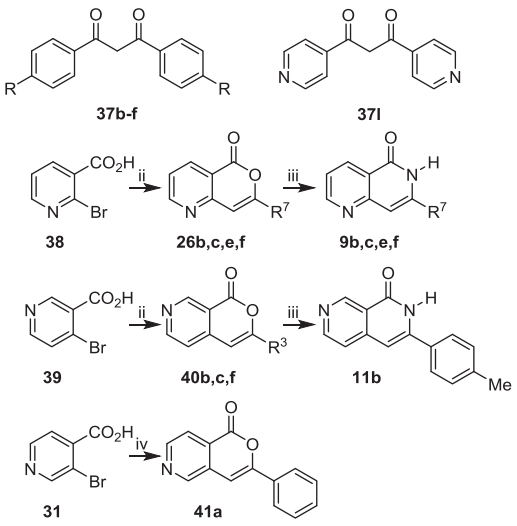
Scheme 1. Synthetic approaches to target arylnaphthyridinones **9**–**11** by acid-catalysed cyclisation of arylethynylpyridinenitriles, N-oxidation to **13**, methylation to **12**, **29a**, **36c** and reduction to the 7-aryl-1,2,3,4-tetrahydro-1,6-naphthyridin-5-ones **17**. Reagents and conditions: (i) BnBr, Cs_2CO_3, DMF; (ii) $Me_3SiC{\equiv}CH$, $(Ph_3P)_2PdCl_2$, CuI, Na ascorbate, Et_3N, THF, Ar, 40 °C; (iii) Bu_4NF, THF; (iv) AcOH, reflux; (v) THF, H_2O, reflux; (vi) Bu_4NBr, P_2O_5, PhMe, reflux; (vii) **21**, $(Ph_3P)_2PdCl_2$, CuI, Na ascorbate, THF, Pr^i_2NH, Ar, 40 °C; (viii) aq H_2SO_4 (9 M), reflux; (ix) NH_3, $MeO(CH_2)_2OH$, pressure, 100 °C; (x) MeI, DMF; (xi) urea·H_2O_2, $(F_3CCO)_2O$, DMF, 0 °C; (xii) BH_3·pyridine, HCO_2H; (xiii) $LiNPr^i_2$, solid CO_2, THF, Ar, −78 °C; (xiv) EtO_2CCl, Et_3N, THF, 0 °C, then NH_3; (xv) $POCl_3$, reflux.

change in chemical shift of 1-N upon oxidation, compared with the small change for 6-N, confirms that oxidation took place at 1-N.

Oxidation of 3-phenyl-3,4-dihydro-2,7-naphthyridin-1-one (from acid-catalysed cyclisation of 3-cyano-4-(2-phenylethenyl)-pyridine) is reported to afford **11a**.[41] In the present work, the 3-aryl-2,7-naphthyridin-1-ones **11** were approached through similar acid-catalysed cyclisations of arylethynylpyridinenitriles. Sonogashira couplings of 4-bromo-3-cyanopyridine **27** with the range of arylethynes **21a**,**c**–**e**,**g** gave the required intermediates **28a**,**c**–**e**,**g** (Scheme 1) in moderate-to-good yields, in the presence of sodium ascorbate. Attempted couplings of **21b** and **21l** with **27** failed, despite the activation of the 4-bromopyridine component by the ring-N. Cyclisation by reaction of **28a**,**c** with hot aqueous sulfuric acid and treatment of the mixtures of 2,7-naphthyridin-1-ones and pyrano[3,4-*c*]pyridin-1-ones with ammonia under pressure gave the required 3-aryl-2,7-naphthyridin-1-ones **11a**,**c**. Curiously, although the major components of the product mixtures from the cyclisations of **28a**,**c** with sulfuric acid were the pyrano[3,4-*c*]pyridin-1-ones, the corresponding acid-catalysed reaction of the 4′-aminophenyl compound **28g** gave only the naphthyridinone **11g**, obviating the need for treatment with ammonia. One example, **11a**, was methylated at 7-N to provide the cation **29a** but all attempts to reduce this to 3-phenyl-5,6,7,8-tetrahydro-2,7-naphthyridin-1-one, by analogy with the preparation of **17**, failed.

Synthesis of synthesise 3-bromo-4-cyanopyridine **30** was required in approaching the 3-aryl-2,6-naphthyridin-1-ones **10a**–**e** in the same way. Lithiation of 3-bromopyridine **30** with LDA and quench of the anion with carbon dioxide gave the 4-carboxylic acid **31** in low yield. Conversion into the amide **32** and dehydration with $POCl_3$ gave the required nitrile **34**. Sonogashira couplings attached the arylalkynes **21a**–**e**, affording **35a**–**e**. Cyclisation and treatment with ammonia furnished the targets **10a**–**c**,**e** but the trifluoromethylphenylalkyne in **35d** was insufficiently nucleophilic to react. Only one example, **10c**, could be methylated at 6-N (affording **36c**) and this cation could not be reduced.

Scheme 2 shows development of an alternative strategy to access the target arylnaphthyridinones. We previously reported a one-pot tandem Hurtley–retro-Claisen-cyclisation sequence to assemble isocoumarins from 2-bromo-3-nitrobenzoic acid and symmetrical and unsymmetrical β-diketones.[42] This was adapted to provide those arylnaphthyridinones which had been inaccessible or accessible only with difficulty by the route shown in Scheme 1. In view of the difficulties experienced in separating mixtures of isocoumarins generated from unsymmetrical β-diketones,[42] only symmetrical 1,3-diarylpropane-1,3-diones **37** were used in the present study. In contrast to the successful Hurtley couplings with 2-bromo-3-nitrobenzoic acid and with 3-bromothiophene-4-carboxylic acid,[42,43] only one example, **31**, gave a usable yield of the corresponding pyranopyridinone **41a**, with **37a** and CuI in boiling DMF. Liu et al. recently published[44] a transition-metal-free variant of the Hurtley reaction, in which 2-bromobenzoic acids are heated with β-diketones in the presence of Cs_2CO_3 in acetonitrile; the S_NAr reaction is followed by retro-Claisen cleavage and cyclisation to form isocoumarins. Adapting this procedure to our target naphthyridinones, 2-bromopyridine-3-carboxylic acid **38** was treated with excesses of the symmetrical β-diketones **37b**,**c**,**e**,**f** and Cs_2CO_3 in boiling acetonitrile to give excellent yields of the corresponding 7-arylpyrano[4,3-*b*]pyridin-5-ones **26b**,**c**,**e**,**f**. The reaction failed for the electron-deficient β-diketones **37d**,**k**, even when the reaction temperature was increased by the use of boiling propionitrile as solvent. The C–Br bond is activated by the adjacent nitrogen in **38**; it is also activated but to a lesser extent in 4-bromopyridine-3-carboxylic acid **39**. Correspondingly, **39** reacted with the β-diketones **37b**,**c**,**f** to give the 3-arylpyrano[3,4-



Scheme 2. Syntheses of pyranopyridinones **26**, **40**, **41** and naphthyridinones **9**, **11** via Hurtley couplings or transition metal-free S_NAr reactions. Reagents and conditions: (i) NaH, THF, Ar, 0 °C, then reflux; (ii) **37**, Cs_2CO_3, MeCN, reflux; (iii) NH_3, $MeO(CH_2)_2OH$, pressure, 100 °C; (iv) **37a**, CuI, K_3PO_4, DMF, Ar, 100 °C.

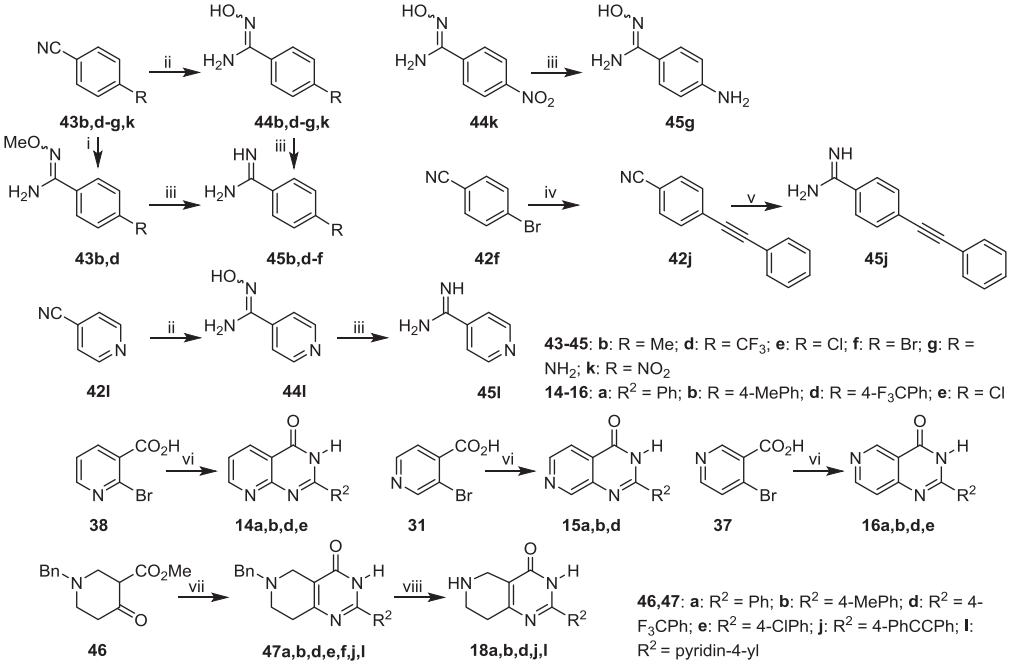
c]pyridin-1-ones **40b**,**c**,**f**. Yields were lower, especially for **40f**, corresponding to the need for a strongly nucleophilic electron-rich enolate. 3-Bromopyridine-4-carboxylic acid **31**, in which the C–Br bond is unactivated, failed to react with any β-diketone, even under forcing conditions. The arylpyranopyridinones **26b**,**c**,**e**,**f**, **40b**,**c**,**f** were converted to the arylnaphthyridinones **9b**,**c**,**e**,**f**, **11b**,**c**,**f** with ammonia in 2-methoxyethanol at elevated temperature and pressure.

Scheme 3 shows the synthetic approaches to the arylpyridinopyrimidinones. The most widely used route to the carbocyclic analogues, the 2-arylquinazolin-4-ones, is acylation of appropriately substituted anthranilamides at the amine nitrogen and base-catalysed cyclisation.[4] However, in transferring this method to the arylpyridinopyrimidinones, all attempts to acylate aminopyridinecarboxamides with benzoyl chlorides failed. Liu et al. have reported an alternative access to quinazolinones through copper-catalysed reaction of benzamidines with 2-bromobenzoic acids.[45] Adapting this method to our targets would need a series of 4-substituted benzamidines **45** to provide diversity.

Following the route of Anbazhagan et al.,[46] attempts were made to convert the readily available 4-substituted benzonitriles **42** into the 4-substituted *N*-methoxybenzamidines **43**, prior to reduction of the N–O bond, but the initial addition reaction failed for all but benzonitrile **42a** and the electron-poor 4-trifluoromethylbenzonitrile **42d**. However, the benzonitriles did react well with hydroxylamine to give the *N*-hydroxyamidines **44** in excellent yields for the electron-neutral and electron-deficient examples **42b**,**d**–**g**,**k**,**l** but failed wholly or partly for the less electrophilic compounds **42c** (0% yield) and **42g** (15%). Transfer hydrogenolysis using ammonium formate and Pd/C (Pt/C for the halo analogues **44e**,**f**) furnished the amidines **45a**,**b**,**d**–**g**,**l**, with the nitro group of **44k** being reduced simultaneously to afford **45g**.

Cu(I)-catalysed cyclocondensation of benzamidines **45a**,**b**,**d** with 2-bromopyridine-3-carboxylic acid **38** afforded moderate yields of 2-arylpyrido[2,3-*d*]pyrimidin-4-ones **14a**,**b**,**d** and a low yield of the chloro analogue **14e** but the reaction failed for **45f**,**g**,**l**. Similar reactions of **45** with 4-bromopyridine-3-carboxylic acid **37**, where the C–Br is also activated, also gave good yields of **16a**,**b**,**d** and a low yield of **16e** and failed to provide **16f**,**g**,**l**. The isomeric 3-bromopyridine-4-carboxylic acid **31** was much less reactive, furnishing only **15a**,**b**,**d** in poor yields.

The amidines were also used in condensations with the cyclic β-keto ester **46** to provide the intermediate 6-benzyl-protected 2-aryl-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidin-4-ones **47**. Since



Scheme 3. Synthetic approaches to arylpyridopyrimidinones **14**–**16** and 2-aryl-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidin-4-ones **18**. Reagents and conditions: (i) $MeONH_2{\cdot}HCl$, Na_2CO_3, MeOH, H_2O (sonication for **42d**); (ii) $NH_2OH{\cdot}HCl$, $NaHCO_3$, EtOH, H_2O, reflux; (iii) $HCOONH_4$, Pd/C (Pt/C for **46e**, **f**), AcOH, Ar, reflux; (iv) **21a**, $(Ph_3P)_4Pd$, CuI, Na ascorbate, Pr^i_2NH, THF, Ar, 40 °C; (v) NH_4Cl, NaOMe, MeOH, reflux; (vi) **45**, CuI, Cs_2CO_3, DMF, Ar, 80 °C; (vii) **45**, NaOMe, MeOH, reflux; (viii), HCOOH, Pd/C, MeOH.

the extended analogue **18j** was also required, 4-phenylethynylbenzamidine **45j** was prepared in two steps (Sonogashira coupling of **42f** with phenylethyne and reaction of the intermediate **42j** with NaOMe and NH_4Cl). Condensation with **46** formed **47j** in good yield. From **47a**,**b**,**d**,**g**, the *N*-benzyl protection was removed by transfer hydrogenolysis under acidic conditions to give the target 2-aryl-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidin-4-ones **18a**,**b**,**d**,**g** in good yields. Interestingly, the alkyne of **47j** passed through this process unscathed to give **18j**.

2.3. Biochemical evaluation

Compounds were each initially screened at three different concentrations for inhibition of the catalytic activity of human TNKS-2 (automodification), using an assay previously developed by us.[3,4] The data are expressed in Table 1 as percentage inhibition of enzyme activity, relative to control (no inhibitor). IC_{50} values were measured for some selected analogues showing more potency in the initial screen and for examples from each series (Table 2). These selected examples were also evaluated for inhibition of TNKS-1 (co-substrate = histone-1) and counterscreened for isoform-selectivity using a PARP-1 inhibition assay, both as previously described.[3,4]

2.4. Structural studies

Compounds **12b** and **17a** were crystallised with TNKS-2 to rationalise the design of inhibitors and to understand their mode of binding (PDB codes 4W5I and 4UX4, respectively). Although TNKS-2 was used in the crystallographic studies, the catalytic domains of TNKS-1 and TNKS-2 are highly homologous and the residues at the inhibitor-binding sites are completely conserved. TNKS-2-inhibitor complexes were crystallised containing two monomers in the asymmetric unit. As the two monomers are almost identical, the discussion will refer to monomer A in both of the structures.

As expected, **12b** and **17a** bind to the nicotinamide-binding site of TNKS-2. The binding mode is typical of the PARP inhibitors, having the H-bonds to Gly^{1032} and Ser^{1068} and the canonical stacking interaction with Tyr^{1071}. This binding is shown in Figure 4 (B and C, respectively), with the corresponding structure of the isoquinolin-1-one **3** ($R^{4'}$ = Me, R^5 = OMe) for comparison (Fig. 4A).[3] The binding mode of the compounds is almost identical, despite the differences in the *para*-substitution on the phenyl ring and the saturation state and charge of the heterocyclic system. When compared with the TNKS-2-nicotinamide complex (PDB code 3U9H),[29] both **12b** and **17a** induce a change in the side-chain of Phe^{1035}, which moves ~1.1 Å towards the *para*-position of the phenyl ring of the compounds. This may reflect the occupation of the hydrophobic cavity by the (substituted)phenyl of **12b** and **17a**, whereas this group is absent from the nicotinamide unit in NAD^+.

2.5. Structure–activity relationships

Examining firstly the 7-aryl-1,6-naphthyridin-5-ones **9**, it is clear that the replacement of carbon-5 of the 5-arylisoquinolin-1-ones **3** with nitrogen has a markedly deleterious effect on binding and potency. For example, **9a**, with a simple phenyl as the aryl group occupying the hydrophobic cavity, shows IC_{50} = 5 μM against TNKS-2 (Table 2), whereas the isoquinolinone analogue **3** ($R^{4'}$ = H, R^5 = Me) has IC_{50} = 1.8 nM,[4] indicating a loss of potency of some 1000-fold. The data from the initial three-concentration screen (Table 1) confirm that there is similar loss of activity (relative to **3**) for analogues with comparable *para*-substitution on the aryl group. The nature of the *para*-substituent is relatively unimportant within this series, with polar groups (in **9g**,**i**) conveying similar activity to non-polar groups (in **9b**,**e**,**f**). Interestingly, small electron-donating and electron-neutral groups are tolerated equally but an electron-withdrawing group (in **9d**) and a slightly longer group (in **9c**) reduce binding somewhat (Table 1). Moving the nitrogen to different positions in the naphthyridinone ring in **10** and **11** gives inhibitors of similar potency to **9** (Tables 1 and 2), that is, 1000-fold loss of potency compared with the carbocyclic analogues **3**. In both **10** and **11**, the nature of the *para*-substituent on the phenyl is unimportant. Quaternisation of the ring-nitrogen in **12**, placing the methyl at the same position as in the highly potent 8-methylquinazolinones **2** and the 5-methylisoquinolinones **3** (R^5 = Me), led to complete inactivity, except in the case of the 4-methylphenyl example **12b**. Similarly, occupying the small binding pocket with oxygen in the N-oxide **13a** also diminished activity, although again the 4-methylphenyl analogue **13b** retained some binding. Moving the quaternised $^+$N–Me cation to other positions in the ring, in **29a** and **36c**, abolished activity. This is unsurprising on steric grounds, as the methyl groups in these latter two compounds would clash severely with the protein surface in this part of the binding site; compounds with substituents in this region are very poor inhibitors of many of the PARP isoforms.

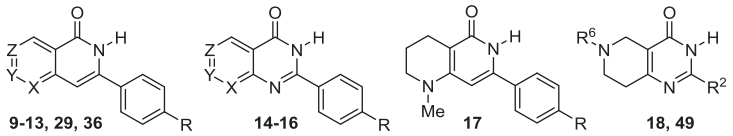
Just as **9**–**11** are aza-analogues of the isoquinolinones **3**, pyridinopyrimidinones **14**–**16** are the aza-analogues of the quinazolinones **2**. There is also an approximately 1000-fold loss in potency in introducing the ring-nitrogen in these series (Table 1), compared with the parent quinazolinones **2**.[3]

Inhibition of the isoform TNKS-1 by **9a** and **10a** broadly followed the activity against TNKS-2 (Table 2). This is unsurprising, as their NAD^+-binding domains have high homology. However, **11b** showed 17-fold selectivity for inhibition of TNKS-1 versus TNKS-2 and is thus one of very few compounds to show this selectivity,[2] although several show selectivity towards TNKS-2.[3]

The dramatic loss in activity of the arylnaphthyridinones **9**–**11** and the arylpyridopyrimidinones **14**–**16**, compared with their carbocyclic equivalents **2** and **3**, appears to be consequent to electron-deficiency. Whereas the potency is diminished 1000-fold for the uncharged analogues **9**–**11**, **14**–**16**, relative to **2**, **3**, it is reduced further for the cationic compounds **12 a**,**c**–**g**. This observation is again consistent with electron-deficiency being deleterious to binding. A rationalisation may be an unfavourable interaction between the electron-poor rings and the positively charged side-chain ammonium of Lys^{1067}. As shown in crystal structures, this ammonium nitrogen is located only 5.5–6.0 Å from 1-N, 2-C and 3-C in **12b** (present work), whereas it is similarly located in the structure of the complex of **3** ($R^{4'}$ = Me, R^5 = OMe) to make an attractive interaction with the electron-rich carbocyclic ring of this latter ligand (Fig. 4).[3] By contrast, the partly-saturated tetrahydropyridine rings of **17a**–**f**, active inhibitors of TNKS-2, are electron-rich and may also make attractive interactions with Lys^{1067} (Fig. 4).

Interestingly, the 7-aryl-1,2,3,4-tetrahydro-1,6-naphthyridin-5-ones **17** showed good inhibitory activity (Table 2). Compounds **11a**,**c**–**f** all inhibited TNKS-2 with IC_{50} < 100 nM, with **17c**,**d** being more potent than XAV939 **1**.[3] In this series, the *para*-substituent on the exocyclic phenyl ring had a marked effect on potency and isoform-selectivity The parent **17a** had weaker and non-selective activity between the tankyrases. Analogues **17c** and **17d** (carrying OMe and CF_3 groups, respectively) were highly potent against both isoforms. Although the *para*-halo compounds **17e** and **17f** retained good potency against TNKS-2, they were much less active against TNKS-1, with **17f** being a potentially useful isoform-selective inhibitor of TNKS-2 (ca. 70-fold selectivity). The structural basis of the marked dependence of potency and selectivity on the nature of this *para*-substituent is unclear. What is clear is the close similarity of the conformation of **17**, as bound into TNKS-2, and the bound conformation of XAV939 **1**. Figure 5 shows **1** and **17a** as bound into TNKS-2, with the protein removed from the structure. This

Table 1
Initial screen of test compounds



Compd	X,Y,Z[a]	R	100 μM	10 μM	1.0 μM	100 nM	10 nM
9a	X = N	H			42 ± 4	0	0
9b	X = N	Me	97 ± 8[b]	100	39 ± 3		
9c	X = N	OMe	93 ± 19	51 ± 22	0		
9d	X = N	CF_3			14 ± 24	14 ± 10	1 ± 10
9e	X = N	Cl	100	96 ± 26	35 ± 25		
9f	X = N	Br		77 ± 8	47 ± 14	25 ± 4	
9g	X = N	NH_2		96 ± 3	74 ± 5	0	
9i	X = N	OH		96 ± 5	58 ± 6	3 ± 13	
10a	Z = N	H			71 ± 8	18 ± 8	0
10b	Z = N	Me		85 ± 6	52 ± 20	0	
10c	Z = N	OMe			78 ± 3	31 ± 11	0
10g	Z = N	NH_2			34 ± 15	14 ± 20	0
11b	Y = N	Me		92 ± 4	51 ± 8	13 ± 10	
11c	Y = N	OMe		97 ± 1	50 ± 13	9 ± 15	
11e	Y = N	Cl		84 ± 7	37 ± 15	0	
12a	X = N^+–Me	H			0	24 ± 12	4 ± 10
12b	X = N^+–Me	Me	100	100	83 ± 12		
12c	X = N^+–Me	OMe		12 ± 5	9 ± 5	0	
12d	X = N^+–Me	CF_3			24 ± 4	0	
12e	X = N^+–Me	Cl		9 ± 6	8 ± 10	17 ± 9	
12f	X = N^+–Me	Br		45 ± 24	27 ± 5	23 ± 1	
12g	X = N^+–Me	NH_2		0	0	0	
13a	X = N^+–O^-	H			23 ± 13	1 ± 6	0
13b	X = N^+–O^-	Me		100	94 ± 2	12 ± 15	
14a	X = N	H	0	0	0		
14b	X = N	Me	71 ± 16	37 ± 20	17 ± 35		
14d	X = N	CF_3		43 ± 9	20 ± 9	21 ± 37	
14e	X = N	Cl		15 ± 3	0	0	
15a	Z = N	H		80 ± 14	29 ± 6		
15b	Z = N	Me		93 ± 7	18 ± 18	0	
15d	Z = N	CF_3		57 ± 6	19 ± 13	0	
16a	Y = N	H		15 ± 9	20 ± 4	0	
16b	Y = N	Me	86 ± 1	62 ± 11	43 ± 19		
16d	Y = N	CF_3		65 ± 5	17 ± 14	0	
16e	Y = N	Cl		81 ± 9	9 ± 6	9 ± 4	
17a		H			99 ± 4	78 ± 3	43 ± 9
17c		OMe		100	82 ± 10	93 ± 15	
17d		CF_3		95 ± 3	86 ± 5	82 ± 19	
17e		Cl		91 ± 14	89 ± 11	24 ± 12	
17f		Br		97 ± 3	89 ± 6	49 ± 7	
18a		R^2 = Ph, R^6 = H		11 ± 14	0	0	
18b		R^2 = 4-MePh, R^6 = H		35 ± 42	0	0	
18d		R^2 = 4-F_3CPh, R^6 = H		7 ± 10	7 ± 11	19 ± 10	
18j		R^2 = 4-PhC≡CPh, R^6 = H		36 ± 7	28 ± 9	0	
18l		R^2 = pyridin-4-yl, R^6 = H		0	0	0	
29a	Z = N^+–Me	H		0	14 ± 14	15 ± 5	
36c	Y = N^+–Me	OMe		0	0	0	
47a		R^2 = Ph, R^6 = Bn		0	0	0	
47b		R^2 = 4-MePh, R^6 = Bn		0	0	0	
47d		R^2 = 4-F_3CPh, R^6 = Bn		0	0	0	
47e		R^2 = 4-ClPh, R^6 = Bn			18 ± 5	0	
47f		R^2 = 4-BrPh, R^6 = Bn		0	0	0	
47j		R^2 = 4-PhC≡CPh, R^6 = Bn		17 ± 15	7 ± 8	29 ± 20	
47l		R^2 = pyridin-4-yl, R^6 = Bn		39 ± 9	0	0	

Percentage inhibition of the catalytic activity of TNKS-2 by arylnaphthyridinones, aryltetrahydronaphthyridinones, arylpyridopyrimidinones and aryltetrahydropyridopyrimidinones.

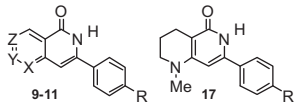
[a] Positions not indicated are CH.

[b] Unoccupied cells = not determined.

illustrates that both compounds adopt the same out-of-plane deformation of the (partly) saturated ring.

Compounds **18** and **47** are aza analogues of **1**, where the sulfur is replaced by a secondary amine (**18**) or a bulky tertiary amine (**47**). Both are strongly basic and would be expected to be protonated and cationic at physiological pH. As expected, the bulk of the benzyl group in this position in **47** abolishes binding and inhibitory activity (Table 1). Less expectedly, **18a**,**b**,**d**,**l** are also devoid of activity against TNKS-2, with **18j** (with the side-chain extension) showing only slight inhibition (36%) at 10 μM.

Table 2
IC_{50} values determined for inhibition of tankyrase-1, tankyrase-2 and PARP-1



Compd	X,Y,Z[a]	R	Tankyrase-1 IC_{50} (nM)	Tankyrase-2 IC_{50} (nM)	PARP-1 IC_{50} (nM)
1	[XAV939]		15 ± 4	15 ± 3	[b]
9a	X = N	H	$10.6 \times 10^3 \pm 6.9 \times 10^3$	$5 \times 10^3 \pm 1.1 \times 10^3$	
10a	Z = N	H	$2.2 \times 10^3 \pm 0.2 \times 10^3$	$1.3 \times 10^3 \pm 0.4 \times 10^3$	
11b	Y = N	Me	350 ± 140	$6.2 \times 10^3 \pm 0.4 \times 10^3$	$43 \times 10^3 \pm 2 \times 10^3$
17a		H	189 ± 1	97 ± 35	949 ± 217
17c		OMe	1.7 ± 1.2	1.1 ± 0.4	$3.4 \times 10^3 \pm 1.0 \times 10^3$
17d		CF_3	7.5 ± 1.9	1.5 ± 0.4	$4.8 \times 10^3 \pm 1.6 \times 10^3$
17e		Cl	134 ± 36	21 ± 1.4	2×10^3
17f		Br	423 ± 256	6.1 ± 0.7	2×10^3

[a] Positions not indicated are CH.
[b] Unoccupied cells = not determined.

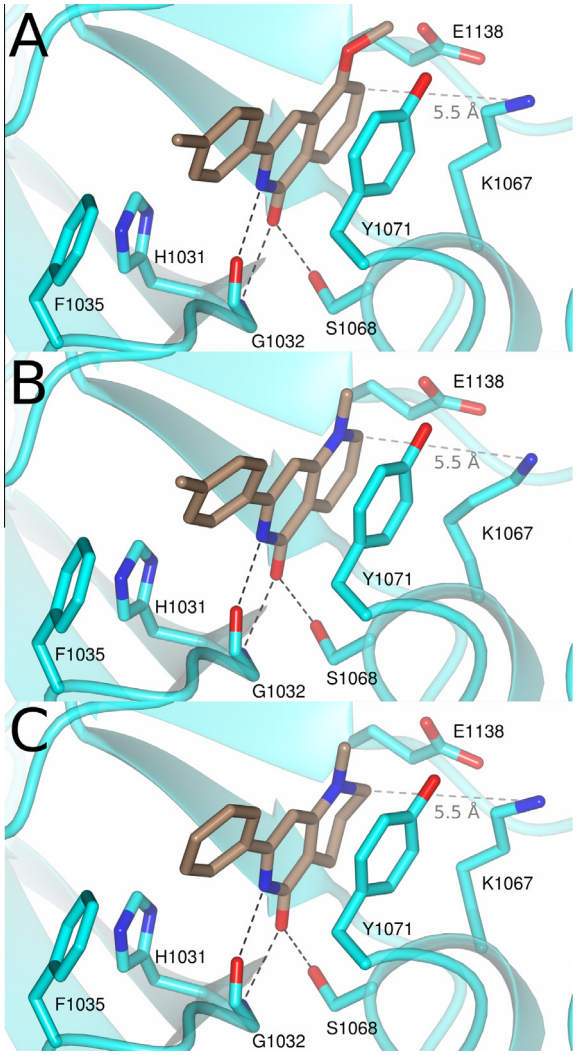


Figure 4. Crystal structures of **12b** (B) and **17a** (C) bound into the nicotinamide-binding site and adjacent hydrophobic cavity of tankyrase-2. The corresponding structure of 5-methoxy-3-(4-methylphenyl)isoquinolin-1-one 3 ($R^{4'}$ = Me, R^5 = OMe) bound into tankyrase-2 is shown for comparison (A).[3] All three structures show the classical H-bonding motif of the lactam to Gly^{1032} and Ser^{1068} and the proximity of the benzene (A), pyridinium (B) and tetrahydropyridine rings of the inhibitors to the ε-N^+H_3 of Lys^{1067}.

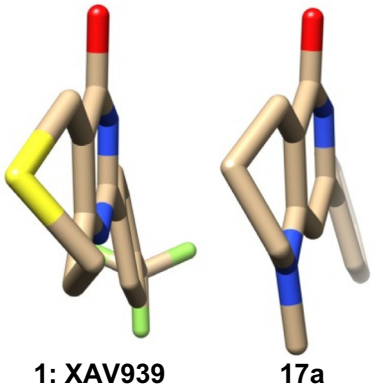
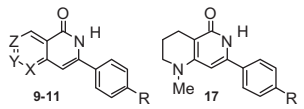


Figure 5. Views of the structures of XAV939 **1** and **17a** as bound into tankyrase-2, showing similarity in conformation.

Compound **18d** is the strict analogue of **1**, so the >1000-fold loss of activity is remarkable. Given the close similarity in structure between **18** and **1**, the loss of activity cannot be due to conformational issues or to steric clash. The major difference between **1** and **18** is that the neutral non-polar sulfur in the former is replaced by a polar cationic entity in the latter. The buckling of the saturated ring of **1** (Fig. 5) places the sulfur in a relatively hydrophobic region, bent away from the ammonium of Lys^{1067}.[25] Here the S is located 3.7 Å from the methyl of Ala^{1062} and immediately above the π-bond of the backbone amide carbonyl of Phe^{1061}, placing it 4.2 Å from the γ-CH_2 of Lys^{1067}. The protonated cationic amine of **18** cannot make favourable interactions in this region. One may speculate that this unfavourable interaction is overcome to some extent by additional binding from the rigid linear phenylethynyl-phenyl unit in **18j** to give the observed modest inhibition; this extension would project into the narrow tunnel at the distal side of the hydrophobic cavity, by analogy with the potent TNKS-2 inhibitor **3** ($R^{4'}$ = —C≡CPh, R^5 = Me).[3]

The potent inhibitors of TNKS-1 and TNKS-2, **17a**,**c**–**f**, were counterscreened for collateral inhibition of the superfamily member PARP-1 (Table 2). Only weak inhibition was observed in preliminary studies, indicating >1000-fold selectivity for the tankyrases for **17c**,**d**, in line with similar selectivities seen for quinazolinones **2** and isoquinolinones **3**.[3,4]

Table 3
Antiproliferative activities against HT29 human colon carcinoma cells and FEK4 human fibroblasts



Compd no.	X,Y,Z[a]	R	HT29 IC_{50} (μM)	FEK4 IC_{50} (μM)
9a	X = N	H	>500[b]	>500[b]
10a	Z = N	H	>500[b]	>500[b]
11b	Y = N	Me	157 ± 88	>500[b]
17a		H	>500[b]	>500[b]
17c		OMe	>200[b]	>200[b]
17d		CF_3	>200[b]	>200[b]
17e		Cl	26 ± 5	>500[b]
17f		Br	26 ± 5	>500[b]

[a] Positions not indicated are CH.
[b] Limited by solubility.

2.6. Antiproliferative activity

Selected analogues showing inhibition of tankyrases were assessed for their antiproliferative activity against HT29 human colon carcinoma cells and against non-malignant FEK4 cells. The latter were isolated by Tyrrell from human foreskin and were a kind gift.[47] They were used as a model for normal tissue. The MTS assay was used, as previously reported by us.[3,4] Most of the compounds examined did not inhibit the proliferation of either cell-type (Table 3). However, **11b** (a weak selective inhibitor of TNKS-1) had slight activity against HT29 cells. More striking was the selective toxicity of the 4′-halophenyl tetrahydronaphthyridinones **17e**,**f** towards the HT29 colon cancer cells, leaving the 'normal' cells unscathed. Isoform-selectivity in inhibition of the tankyrases appears to correlate with selective anticancer cytotoxicity in this very preliminary study but there is, as yet, no evidence of a causal relationship.

3. Conclusions

In this paper, we report the design of series of arylnaphthyridinones and arylpyridopyrimidinones as aza analogues of 3-arylisoquinolin-1-ones **3** and 2-arylquinazolin-4-ones **2** (known potent and selective inhibitors),[3,4] respectively, to test the tolerance of the inhibitor-binding motif of the tankyrases to the additional ring-nitrogen. These compounds were assembled by acid-catalysed cyclisation of the corresponding arylethynylpyridinenitriles to lactones and conversion to the naphthyridinones with ammonia. Methylation and oxidation took place at 1-N of 7-aryl-1,6-naphthyridin-5-ones, to introduce a small group in the region of space occupied by the C–Me in the potent 3-aryl-5-methylisoquinolin-1-ones and 2-aryl-8-methylquinazolin-4-ones.[3,4] However, the electron-deficient pyridine ring in the naphthyridinones (replacing a benzene ring in the potent compounds) severely reduced binding and inhibitory potency, by up to three orders of magnitude. These results are rationalised by examining crystallographically the mode of binding of one analogue, **12b**. This compound makes the expected H-bonds with Gly^{1032} and Ser^{106}, with the benzene ring in the centre of the adjacent hydrophobic cavity; however, binding may be reduced by the proximity of the pyridine/pyridinium to the positively charged side-chain of Lys^{1067}. Examining more flexible analogues, the tetrahydropyridopyrimidines **18** are strict aza-analogues of the known inhibitor XAV939 (**1**)[9,25] but replacement of lipophilic sulfur with (protonated) nitrogen abolished activity, owing to unfavourable interactions with the binding pocket, which is hydrophobic in this region. By contrast the 7-aryl-1-methyltetrahydronaphthyridinones **17** were highly potent for inhibition of the tankyrases. Interestingly, a crystal structure of **17a** bound into TNKS-2 revealed that it adopts the same buckle of the partly-saturated ring as does **1**.[25] Compounds **17** were also shown to be selective for the tankyrases, inhibiting PARP-1 only at μM concentrations.

These studies have allowed more definition of the structure–activity relationships for binding to and inhibiting the tankyrases, rationalised by structural understanding of the mode of binding. The identification of **17** as potent and selective inhibitors provides a new framework for design of improved agents and optimisation of pharmaceutical properties.

4. Experimental

4.1. Chemistry

Chemical reagents were purchased from Sigma Aldrich, Goss Scientific, Alfa Aesar and Fisher Scientific and were used as supplied. ^1H and ^{13}C NMR spectra were recorded at 400.04 MHz or 500.13 MHz (^1H) and 100.59 MHz or 125.76 MHz (^{13}C), using $(CD_3)_2SO$ containing $SiMe_4$, unless otherwise noted. Signals in ^{13}C NMR spectra were assigned using HSQC and HMBC and signals in ^{15}N spectra with HMBC. Reactions were monitored by thin-layer chromatography on silica gel 60 Å (particle size 40–63 μm). MS data were obtained by electrospray ionisation using a microTOF MS (Bruker Daltonics, Germany) and were calibrated using HCO_2Na. Melting points were measured using a heated stage microscope (Reichert-Jung). Experiments were at ambient temperature, unless otherwise noted. Solutions in organic solvents were dried with $MgSO_4$. Experimental methods for the syntheses of **19h**, **20c–e**,**g**,**h**,**l**, **21c–e**,**g**,**l**, **24**, **31–34**, **37b–f**,**l**, **42j**, **43b**,**d**, **44d**,**d–g**,**k**,**l** and **45b**,**d–g**,**j**,**l** can be found in the Supplementary information.

4.1.1. 7-Phenyl-1,6-naphthyridin-5-one (9a)

Compound **25a** (300 mg, 1.5 mmol) was stirred under reflux in aq H_2SO_4 (9 M, 20 mL) for 1 h. After cooling, aq NaOH (5 M) was added to pH 9. The mixture was extracted with EtOAc (5 × 25 mL). The combined extracts were dried. The evaporation residue was transferred into a pressure tube and dissolved in $MeO(CH_2)_2OH$ (10 mL). NH_3 was passed through the solution until it was saturated, the vessel was closed and the mixture was heated at 100 °C for 30 min. The mixture was cooled in ice and NH_3 was passed through again, followed by closure and heating (30 min). The cycle was repeated twice. Evaporation and recrystallisation $(MeO(CH_2)_2)OH$ gave **9a** (249 mg, 75%) as a white solid: mp 226–230 °C (lit.[30] mp 228–229 °C); ^1H NMR δ 6.98 (1H, s, 8-H), 7.54 (1H, dd, J = 8.4, 4.9 Hz, 3-H), 7.57–7.60 (3H, m, Ph 3,4,5-H_3), 7.89 (2H, d, J = 7.9 Hz, Ph 2,6-H_2), 8.57 (1H, dd, J = 8.0, 1.3 Hz, 4-H), 8.98 (1H, dd, J = 4.5, 1.8 Hz, 2-H), 11.74 (br, NH); ^{13}C NMR δ 104.56 (8-C), 120.40 (3-C), 121.62 (4a-C), 126.99 (Ph 3,5-C_2), 128.82 (Ph 2,6-C_2), 129.82 (Ph 4-C), 133.36 (Ph 1-C), 135.00 (4-C), 143.97 (7-C), 154.16 (2-C), 154.97 (8a-C), 162.93 (5-C); ^{15}N NMR ($(CD_3)_2SO$) δ 124.89 (6-N), 278.87 (1-N), MS m/z 245.0676 $(M+Na)^+$ ($C_{14}H_{10}N_2NaO$ requires 245.0691).

4.1.2. 7-(4-Methylphenyl)-1,6-naphthyridin-5-one (9b)

Compound **25b** was treated with H_2SO_4, then NH_3, as for the synthesis of **9a**, to give **9b** (48%) as an off-white solid: mp 255–258 °C; ^1H NMR δ 2.43 (3H, s, Me), 6.95 (1H, s, 8-H), 7.38 (2H, d, J = 8.0 Hz, Ph 3,5-H_2), 7.54 (1H, dd, J = 8.0, 4.5 Hz, 3-H), 7.79 (2H, d, J = 8.2 Hz, Ph 2,6-H_2), 8.55 (1H, ddd, J = 8.0, 1.6, 0.7 Hz, 4-H), 8.97 (1H, dd, J = 4.5, 1.8 Hz, 2-H), 11.74 (1H, br, NH); ^{13}C NMR δ 20.81 (Me), 103.99 (8-C), 120.27 (4a-C), 121.43 (3-C), 126.82 (Ph 2,6-C_2), 129.38 (Ph 3,5-C_2), 130.54 (Ph 1-C), 134.97 (4-C), 139.60

(Ph 4-C), 144.01 (7-C), 154.27 (8a-C), 154.93 (2-C), 162.94 (5-C); MS *m*/*z* 259.0826 $(M+Na)^+$ ($C_{15}H_{12}N_2NaO$ requires 259.0847).

4.1.3. 7-(4-Methoxyphenyl)-1,6-naphthyridin-5-one (9c)

Compound **25c** was treated with H_2SO_4, then NH_3, as for the synthesis of **9a**, to give **9c** (70%) as an off-white solid: mp 276–280 °C; ^{1}H NMR δ 3.89 (3H, s, Me), 6.92 (1H, s, 8-H), 7.12 (2H, d, J = 8.9 Hz, Ph 3,5-H_2), 7.50 (1H, dd, J = 8.0, 4.6 Hz, 3-H), 7.86 (2H, d, J = 8.9 Hz, Ph 2,6-H_2), 8.54 (1H, dd, J = 8.0, 1.8 Hz, 4-H), 8.95 (1H, dd, J = 4.5, 1.8 Hz, 2-H), 11.65 (1H, br, NH); ^{13}C NMR δ 55.36 (Me), 114.24 (Ph 3,5-C_2), 120.01 (8-C), 122.42 (4a-C), 123.65 (Ph 1-C), 125.60 (3-C), 128.39 (Ph 2,6-C2), 134.94 (7-C), 136.92 (4-C), 143.77 (2-C), 154.90 (8a-C), 156.99 (5-C), 158.82 (Ph 4-C); MS *m*/*z* 275.0767 $(M+Na)^+$ ($C_{15}H_{12}N_2NaO_2$ requires 275.0796), 253.0978 $(M+H)^+$ ($C_{15}H_{13}N_2O_2$ requires 253.0977).

4.1.4. 7-(4-Trifluoromethylphenyl)-1,6-naphthyridin-5-one (9d)

Compound **25d** was treated with H_2SO_4, then NH_3, as for the synthesis of **9a**, to give **9d** (55%) as a white solid: mp >325 °C; ^{1}H NMR δ 7.03 (1H, s, 8-H), 7.53 (1H, dd, J = 8.0, 4.6 Hz, 3-H), 7.92 (2H, d, J = 8.4 Hz, Ph 3,5-H_2), 8.10 (2H, d, J = 8.2 Hz, Ph 2,6-H_2), 8.53 (1H, dd, J = 8.0, 1.7 Hz, 4-H), 9.00 (1H, dd, J = 4.5, 1.8 Hz, 2-H), 11.25 (1H, br, NH); ^{13}C NMR 105.92 (8-C), 120.90 (4a-C), 122.13 (3-C), 124.1 (q, J = 264 Hz, CF_3), 125.62 (q, J = 3.6 Hz, Ph 3,5-C_2), 128.03 (Ph 2,6-C_2), 129.78 (q, J = 32 Hz, Ph 4-C), 135.05 (4-C), 137.31 (Ph 1-C), 142.51 (7-C), 153.90 (2-C), 155.09 (8a-C), 162.86 (5-C); ^{15}N NMR δ 123.4 (6-N), 279.7 (1-N); ^{19}F NMR δ −61.20 (s, CF_3); MS *m*/*z* 291.0727 $(M+H)^+$ ($C_{15}H_{10}N_2O$ requires 291.0745).

4.1.5. 7-(4-Chlorophenyl)-1,6-naphthyridin-5-one (9e)

Compound **25e** was treated with H_2SO_4, then NH_3, as for the synthesis of **9a**, to give **9d** (33%) as an off-white solid: mp 296–298 °C; IR ν_{max} 3163, 1669, 1627, 1588, 720 cm^{-1}; ^{1}H NMR δ 6.95 (1H, s, 8-H), 7.50 (1H, dd, J = 8.0, 4.5 Hz, 3-H), 7.58 (2H, d, J = 9.0 Hz, Ph 3,5-H_2), 7.91 (2H, d, J = 8.5 Hz, Ph 2,6-H_2), 8.51 (1H, d, J = 8.0 Hz, 4-H), 8.98 (1H, d, J = 4.5 Hz, 2-H), 11.74 (1H, br, NH); ^{13}C δ 104.95 (8-C), 120.58 (4a-C), 121.83 (3-C), 128.82 (Ph 3,5-C_2), 128.93 (Ph 2,6-C2), 134.60 (4-C), 135.03 (Ph 1-C), 155.05 (C=O), 162.90 (8a-C); ^{15}N NMR (HMBC) δ 124.20 (s, 6-N); MS *m*/*z* 279.0278 $(M+Na)^+$ ($C_{14}H_9{}^{35}ClN_2NaO$ requires 279.0296), 259.0420 $(M+H)^+$ ($C_{14}H_{10}{}^{37}ClN_2O$ requires 259.0450), 257.0482 $(M+H)^+$ ($C_{14}H_{10}{}^{35}ClN_2O$ requires 257.0476).

4.1.6. 7-(4-Bromophenyl)-1,6-naphthyridin-5-one (9f)

NH_3 was bubbled through **26f** (140 mg, 0.47 mmol) in $MeO(CH_2)_2OH$ (10 mL) for 10 min in a pressure vessel. The vessel was closed and the mixture was heated at 120 °C for 30 min. The reaction mixture was cooled in ice and NH_3 was bubbled again, followed by closure and heating (30 min). The cycle was repeated twice. Evaporation and recrystallisation (EtOH) gave **9f** (100 mg, 75%) as a buff powder: mp 304–305 °C; ^{1}H NMR δ 7.05 (1H, s, 8-H), 7.50 (1H, dd, J = 7,7, 4.0 Hz, 3-H), 7.39 (2H, d, J = 8 Hz, Ph 3,5-H_2), 7.84 (2H, d, J = 8 Hz, Ph 2,6-H_2), 8.30 (1H, d, J = 7.7 Hz, 4-H), 8.69 (1H, d, J = 4.8 Hz, 2-H), 9.15 (1H, br, NH); ^{13}C NMR δ 105.80 (8-C), 122.51 (4a-C), 122.97 (Ph 4-C), 123.61 (3-C), 127.41 (Ph 1-C), 130.22 (Ph 2,6-C_2), 130.75 (Ph 3,5-C_2), 131.91 (7-C), 134.61 (4-C), 156.71 (8a-C), 168.09 (5-C); MS *m*/*z* 627/625/623 $(2M+Na)^+$, 324.9771 $(M+Na)^+$ ($C_{14}H_9{}^{81}BrN_2NaO$ requires 324.9774), 322.9790 $(M+Na)^+$ ($C_{14}H_9{}^{79}BrN_2NaO$ requires 322.9805), 302.9964 $(M+H)^+$ ($C_{14}H_{10}{}^{81}BrN_2O$ requires 302.9951), 301.0024 $(M+H)^+$ ($C_{14}H_{10}{}^{79}BrN_2O$ requires 300.9971).

4.1.7. 7-(4-Aminophenyl)-1,6-naphthyridin-5-one (9g)

Compound **26g** was treated with H_2SO_4, then NH_3, as for the synthesis of **9a**, to give **9g** (66%) as a yellow powder: mp >300 °C; ^{1}H NMR δ 5.65 (2H, br, NH_2), 6.80 (1H, s, 8-H), 7.43 (1H, dd, J = 8.0, 4.5 Hz, 3-H), 7.60 (2H, d, J = 8.6 Hz, Ph 3,5-H_2), 7.71 (2H, d, J = 8.7 Hz, Ph 2,6-H_2), 8.49 (1H, dd, J = 8.0, 1.8 Hz, 4-H), 8.90 (1H, dd, J = 4.8, 1.8 Hz, 2-H), 11.50 (1H, br, NH); ^{13}C NMR δ 99.08 (8-C), 113.59 (Ph 3,5-C_2), 117.55 (3-C), 122.16 (4a-C), 124.87 (Ph 1-C), 127.78 (4-C), 130.51 (Ph 2,6-C_2), 136.95 (7-C), 150.62 (2-C), 153.57 (Ph 4-C), 156.59 (8a-C), 161.65 (5-C).

4.1.8. 7-(4-Hydroxyphenyl)-1,6-naphthyridin-5-one (9i)

Compound **25h** was treated with H_2SO_4, then NH_3, as for the synthesis of **9a**, to give **9i** (10%) as a pale buff solid: mp 258–260 °C; IR ν_{max} 3423, 1660, 1617, 1589 cm^{-1}; ^{1}H NMR δ 6.87 (1H, s, 8-H), 6.93 (2H, d, J = 8.6 Hz, Ph 2,6-H_2), 7.37 (1H, d, J = 7.4 Hz, 3-H), 7.73 (2H, d, J = 8.6 Hz, Ph 3,5-H_2), 8.52 (1H, d, J = 2.5 Hz, 4-H), 8.94 (1H, d, J = 2.5 Hz, 2-H), 9.99 (1H, br, OH), 11.67 (1H, br, NH); ^{13}C NMR δ 115.57 (8-C), 119.0 (3-C), 121.0 (Ph 1-C), 126.0 (4a-C), 128.11 (Ph 2,6-C_2), 128.42 (Ph 3,5-C_2), 135.0 (4-C), 147.0 (2-C), 153.0 (Ph 4-C), 154.89 (8a-C), 163.0 (5-C); MS *m*/*z* 239.0816 $(M+H)^+$ ($C_{14}H_{11}N_2O_2$ requires 239.0816); MS *m*/*z* 237.0632 $(M-H)^-$ ($C_{14}H_9N_2O_2$ requires 237.0659).

4.1.9. 3-Phenyl-2,6-naphthyridin-1-one (10a)

Compound **35a** (100 mg, 0.49 mmol) was stirred at 120 °C in polyphosphoric acid (10 mL) for 1 h. After cooling, aq NaOH (5 M) was added to pH 8. The mixture was extracted (EtOAc, 3×). The combined extracts were dried. The evaporation residue was transferred into a pressure tube and dissolved in $MeO(CH_2)_2OH$ (5 mL). NH_3 was bubbled through the solution, the vessel was closed and the mixture was heated at 130 °C for 30 min. The reaction mixture was cooled in ice and NH_3 was bubbled again, followed by closure and heating (30 min). The cycle was repeated thrice. Evaporation and trituration (EtOH) gave an inseparable mixture of **10a** and **32** (85 mg) as a gum. ^{1}H NMR showed that the mixture comprised **10a** (56% yield) and **32** (22% yield). **10a**: ^{1}H NMR δ 7.11 (1H, s, 4-H), 7.39 (1H, m, Ph 4-H), 7.42 (2H, m, Ph 3,5-H_2), 7.67 (2H, d, J = 7.0 Hz, Ph 2,6-H_2), 8.64 (1H, s, 8-H), 8.70 (1H, s, 7-H), 8.99 (1H, s, 5-H), 11.68 (1H, br, NH); ^{13}C NMR δ 101.50 (4-C), 121.20 (8-C), 125.50 (m, Ph 3,4,5-C_3), 128.50 (Ph 2,6-C_2), 131.00 (Ph 1-C), 132.90 (8a-C), 146.50 (7-C), 151.50 (5-C); MS *m*/*z* 223.0875 $(M+H)^+$ ($C_{14}H_{11}N_2O$ requires 223.0866). **32**: ^{1}H NMR δ 7.10 (1H, s, 4-H), 7.86 (2H, d, J = 8.2 Hz, Ph 2,6-H_2), 7.98–8.00 (3H, m, Ph 3,4,5-H_3), 8.08 (1H, d, J = 5.2 Hz, 8-H), 8.67 (1H, d, J = 5.3 Hz, 7-H), 9.17 (1H, s, 5-H); ^{13}C NMR δ 98.52 (4-C), 118.0 (8-C), 125.42 (Ph 2,6-C_2), 127.00 (8a-C), 129.01 (Ph 3,5-C_2), 130.64 (Ph 4-C), 131.20 (Ph 1-C), 132.00 (3-C), 148.44 (7-C), 149.00 (5-C), 151.00 (1-C); MS *m*/*z* 245 (M+Na), 223.0875 $(M+H)^+$ ($C_{14}H_{11}N_2O$ requires 223.0866).

4.1.10. 3-(4-Methylphenyl)-2,6-naphthyridin-1-one (10b)

Compound **35b** was treated with polyphosphoric acid, then with NH_3, as for the synthesis of **10a**, except that the product was recrystallised (EtOH) to give **10b** (6%) as an off-white solid: mp 188–189 °C; ^{1}H NMR δ 7.07 (1H, s, 4-H), 7.38 (2H, d, J = 7.9 Hz, Ph 3,5-H_2), 7.76 (2H, d, J = 8.2 Hz, Ph 2,6-H_2), 8.04 (1H, d, J = 5.3 Hz, 8-H), 8.66 (1H, d, J = 5.3 Hz, 7-H), 9.15 (1H, d, J = 0.8 Hz, 5-H), 11.90 (1H, br, NH); ^{13}C NMR δ 20.70 (Me), 100.10 (4-C), 119.80 (8-C), 127.10 (Ph 2,6-C_2), 129.53 (Ph 3,5-C_2), 146.20 (7-C), 148.90 (5-C); MS *m*/*z* 495.1796 $(2M+Na)^+$ ($C_{30}H_{24}N_4NaO_2$ requires 495.1797), 259.0834 $(M+Na)^+$ ($C_{15}H_{12}N_2NaO$ requires 259.0847), 237.1022 $(M+H)^+$ ($C_{15}H_{13}N_2O$ requires 237.1028).

4.1.11. 3-(4-Methoxyphenyl)-2,6-naphthyridin-1-one (10c)

Compound **35c** was treated with polyphosphoric acid, then NH_3, as for the synthesis of **10b**, to give **10c** (37%) as an off-white solid: mp 244–245 °C; ^{1}H NMR δ 3.90 (3H, s, Me), 7.11 (1H, s, 4-H), 7.12 (2H, d, J = 8.4 Hz, Ph 3,5-H_2), 7.63 (2H, d, J = 8.9 Hz, Ph 2,6-H_2), 7.80 (1H, d, J = 5.1Hz, 8-H), 8.83 (1H, d, J = 5.1Hz, 7-H), 9.04 (1H, s,

5-H); ^{13}C NMR δ 55.54 (Me), 113.87 (Ph 3,5-C_2), 114.74 (4-C), 121.10 (4a-C), 124.89 (8-C), 130.32 (Ph 2,6-C_2), 131.82 (Ph 1-C), 134.20 (8a-C), 135.10 (3-C), 149.01 (7-C), 152.10 (5-C), 160.42 (1-C), 160.99 (Ph 4-C); MS *m*/*z* 527.1679 $(2M+Na)^+$ ($C_{30}H_{24}N_4NaO_4$ requires 527.1695), 275.0772 $(M+Na)^+$ ($C_{15}H_{12}N_2NaO_2$ requires 275.0796), 253.0960 $(M+H)^+$ ($C_{15}H_{13}N_2O_2$ requires 253.0977).

4.1.12. 3-(4-Chlorophenyl)-2,6-naphthyridin-1-one (10e)

Compound **35e** was treated with polyphosphoric acid, then NH_3, as for the synthesis of **10b**, to give **10e** (27%) as an off-white solid: mp 257–259 °C; ^{1}H NMR δ 7.12 (1H, s, 4-H), 7.64 (2H, d, *J* = 8.7 Hz, Ph 3,5-H_2),7.88 (2H, d, *J* = 8.6 Hz, Ph 2,6-H_2), 8.05 (1H, d, *J* = 5.3 Hz, 8-H), 8.69 (1H, d, *J* = 5.3 Hz, 7-H), 9.17 (1H, s, 5-H), 11.75 (1H, br, NH); ^{13}C NMR δ 101.92 (4-C), 119.10 (8-C), 128.73 (Ph 2,6-C_2), 128.82 (Ph 3,5-C_2), 130.01 (4a-C), 132.05 (Ph 1-C), 134.10 (Ph 4-C),137.10 (8a-C), 142.00 (3-C), 146.53 (7-C), 150.20 (5-C), 156.31 (1-C); MS *m*/*z* 279 $(M+Na)^+$, 257.0475 $(M+H)^+$ ($C_{14}H_{10}{}^{35}ClN_2O$ requires 257.0482).

4.1.13. 3-Phenyl-2,7-naphthyridin-1-one (11a)

Compound **28a** (300 mg, 1.5 mmol) was stirred under reflux in aq H_2SO_4 (9 M, 20 mL) for 1 h. After cooling, aq NaOH (5 M) was added to pH 9. The mixture was extracted with EtOAc (5 × 25 mL). The evaporation residue was transferred into a pressure tube and dissolved in $MeO(CH_2)_2OH$ (10 mL). NH_3 was passed through the solution, the vessel was closed and the mixture was heated at 100 °C for 30 min. The reaction mixture was cooled in ice and NH_3 was passed through again, followed by closure and heating (30 min). The cycle was repeated twice. Evaporation and recrystallisation ($MeO(CH_2)_2OH$) gave **11a** (131 mg, 40%) as an off-white solid: mp 236–237 °C (lit.[34] 237–238 °C); IR ν_{max} 3447, 1669, 1631, 1595, 1461 cm^{-1}; ^{1}H NMR δ 6.97 (1H, s, 4-H), 7.58 (3H, m, Ph 3,4,5-H_3), 7.66 (1H, d, *J* = 5.4 Hz, 5-H), 7.86 (2H, dd, *J* = 7.6 Hz, Ph 2,6-H_2), 8.76 (1H, d, *J* = 5.4 Hz, 6-H), 9.38 (1H, s, 8-H), 11.91 (1H, br, NH); ^{13}C NMR δ 101.41 (4-C), 119.66 (5-C), 127.12 (Ph 2,6-C_2), 128.87 (Ph 3,5-C_2), 130.14 (Ph 4-C), 133.26 (Ph 1-C), 143.11 (4a-C), 145.27 (8a-C), 149.84 (8-C), 150.98 (6-C), 162.37 (1-C); MS *m*/*z* 223.0866 $(M+H)^+$ ($C_{14}H_{11}N_2O$ requires 223.0935).

4.1.14. 3-(4-Methylphenyl)-2,7-naphthyridin-1-one (11b)

NH_3 was bubbled through **40b** (35 mg, 0.15 mmol) in $MeO(CH_2)_2OH$ (10 mL) for 10 min in a pressure vessel. The vessel was closed and the mixture was heated at 120 °C for 30 min. The reaction mixture was cooled in ice and NH_3 was bubbled again, followed by closure and heating (30 min). The cycle was repeated twice. Evaporation and recrystallisation (EtOH) gave **11b** (12 mg, 31%) as an off-white powder: mp > 300 °C (decomp.); ^{1}H NMR δ 2.43 (3H, s, Me), 6.94 (1H, s, 4-H), 7.38 (2H, d, *J* = 8.1 Hz, Ph 3,5-H_2), 7.63 (1H, d, *J* = 5.6 Hz, 5-H), 7.77 (2H, d, *J* = 8.2 Hz, Ph 2,6-H_2), 8.73 (1H, d, *J* = 5.5 Hz, 6-H), 9.36 (1H, s, 8-H), 11.70 (1H, br, NH); ^{13}C NMR δ 21.16 (Me), 100.79 (4-C), 119.55 (8a-C), 127.40 (Ph 2,6-C_2,), 128.67 (5-C), 129.38 (Ph 3,5-C_2), 129.40 (Ph 1-C), 144.10 (Ph 4-C), 145.23 (4a-C), 146.00 (3-C), 150.88 (8-C), 153.00 (6-C), 162.36 (1-C); MS *m*/*z* 495 $(2M+Na)^+$, 259.0841 $(M+Na)^+$ ($C_{15}H_{12}N_2NaO$ requires 259.0842).

4.1.15. 3-(4-Methoxyphenyl)-2,7-naphthyridin-1-one (11c)

Compound **28c** was treated with H_2SO_4, then NH_3, as for the synthesis of **11a**, to give **11c** (32%) as a pale orange solid: mp >300 °C (decomp.); ^{1}H NMR δ 3.89 (3H, s, Me), 6.90 (1H, s, 4-H), 7.13 (2H, d, *J* = 8.5 Hz, Ph 3,5-H_2), 7.62 (1H, d, *J* = 8.9 Hz, 5-H), 7.84 (2H, d, *J* = 9.0 Hz, Ph 2,6-H_2), 8.72 (1H, d, *J* = 5.4, 1.3 Hz, 6-H), 9.35 (1H, s, 8-H), 11.82 (1H, br, NH); ^{13}C NMR δ 55.41 (Me), 100.15 (4-C), 114.27 (Ph 3,5-C_2), 119.34 (4a-C), 119.44 (5-C), 125.39 (Ph 1-C), 128.54 (Ph 2,6-C_2), 143.26 (8a-C), 145.02 (3-C), 149.80 (8-C), 150.84 (6-C), 160.78 (Ph 4-C), 162.40 (1-C); MS *m*/*z* 253.0985 $(M+Na)^+$ ($C_{15}H_{13}N_2O_2$ requires 253.0972).

4.1.16. 3-(4-Aminophenyl)-2,7-naphthyridin-1-one (11g)

Compound **28g** (40 mg, 0.18 mmol) was stirred under reflux in aq H_2SO_4 (9 M, 20 mL) for 1 h. After cooling, aq NaOH (5 M) was added to pH 9. The mixture was extracted (EtOAc, 5 × 25 mL). Aq HCl (9 M) was added to the aqueous layer to pH 1. The latter was washed thrice with EtOAc. The solution was basified with aq NaOH (5 M) to pH 9 and extracted with EtOAc (5×). The extracts were dried. Evaporation and recrystallisation (EtOAc) gave **11g** (15 mg, 36%) as a white solid: mp 235–236 °C; IR ν_{max} 3172, 1669, 1617, 1594 cm^{-1}; ^{1}H NMR δ 5.71 (2H, br, NH_2), 6.69 (2H, dd, *J* = 8.8 Hz, Ph 3,5-H_2), 6.77 (1H, s, 4-H), 7.54 (1H, d, *J* = 5.7 Hz, 5-H), 7.59 (2H, d, *J* = 8.8 Hz, Ph 2,6-H_2), 8.65 (1H, d, *J* = 5.4 Hz, 6-H), 9.28 (1H, s, 8-H), 11.74 (1H, br, NH); ^{13}C NMR δ 97.99 (4-C), 112.43 (Ph 1-C), 113.48 (3,5-C_2), 199.10 (5-C), 127.96 (2,6-C_2), 130.41 (3-C), 143.56 (4a-C), 145.99 (8a-C), 150.55 (8-C), 150.91 (6-C), 152.0 (Ph 4-C), 162.46 (1-C); MS *m*/*z* 238.0950 $(M+H)^+$ ($C_{14}H_{12}N_3O$ requires 238.0975).

4.1.17. 1-Methyl-5-oxo-7-phenyl-1,6-naphthyridinium iodide (12a)

MeI (410 mg, 2.9 mmol) was stirred with **9a** (120 mg, 0.54 mmol) in dry DMF (5 mL) for 36 h. The mixture was poured into Me_2CO (3 mL). The solid was collected, washed (Me_2CO) and dried to give **12a** (142 mg, 72%) as a yellow solid: mp 280–284 °C, ^{1}H NMR δ 4.40 (3H, s, Me), 7.18 (1H, s, 8-H), 7.63 (3H, m, Ph 3,4,5-H_3), 7.88 (1H, dd, *J* = 8.0, 6.0 Hz, 3-H), 7.98 (2H, d, *J* = 7.5 Hz, Ph 2,6-H_2), 9.09 (1H, d, *J* = 7.5 Hz, 4-H), 9.20 (1H, d, *J* = 5.5 Hz, 2-H), 12.87 (1H, s, 6-H); ^{13}C NMR δ 44.97 (Me), 93.61 (8-C), 121.24 (3-C), 123.03 (4a-C), 128.05 (Ph 2,6-C_2), 128.98 (Ph 3,5-C_2), 131.55 (Ph 4-C), 132 (Ph 1-C), 144.13 (4-C), 147.98 (8a-C), 150.91, 153 (7-C), 161 (5-C); MS *m*/*z* 237.1012 $(M)^+$ ($C_{15}H_{13}N_2O$ requires 237.1022).

4.1.18. 1-Methyl-7-(4-methylphenyl)-5-oxo-1,6-naphthyridin-1-ium iodide (12b)

Compound **9b** was treated with MeI, as for the synthesis of **12a**, to give **12b** (64%) as a yellow solid: mp 273–276 °C; IR ν_{max} 3431, 3250, 1665, 1606, 1507 cm^{-1}; ^{1}H NMR δ 2.41 (3H, s, PhMe), 4.38 (3H, s, NMe), 7.13 (1H, s, 8-H), 7.40 (2H, d, *J* = 8.0 Hz, Ph 3,5-H_2), 7.82 (1H, m, 3-H), 7.92 (2H, d, *J* = 8.0 Hz, Ph 2,6-H_2), 9.04 (1H, d, *J* = 8.0 Hz, 4-H), 9.16 (1H, br, 2-H); ^{13}C NMR δ 20.99 (PhMe), 44.80 (NMe), 92.55 (8-C), 120.59 (3-C), 122.78 (4a-C), 127.91 (Ph 2,6-C_2), 129.43 (Ph 1-C), 129.52 (Ph 3,5-C_2), 141.69 (Ph 4-C), 143.98 (4-C), 148.05 (8a-C), 150.55 (2-C), 153.5 (br, 7-C); ^{15}N NMR δ 155.00 (1-N); MS *m*/*z* 251.1175 $(M)^+$ ($C_{16}H_{15}N_2O$ requires 251.1179).

4.1.19. 7-(4-Methoxyphenyl)-1-methyl-5-oxo-1,6-naphthyridin-1-ium iodide (12c)

MeI (144 mg, 1.0 mmol) was stirred with **9c** (76 mg, 0.3 mmol) in dry DMF (5 mL) for 3 d. The mixture was poured into EtOAc (25 mL) and petroleum ether (25 mL). The solid was collected, washed (petroleum ether) and dried to give **12c** (5.0 mg, 4%) as a yellow solid: mp >300 °C (decomp.); ^{1}H NMR δ 3.94 (3H, s, OMe), 4.46 (3H, s, NMe), 7.19 (1H, s, 8-H), 7.22 (2H, d, *J* = 8.9 Hz, Ph 3,5-H_2), 7.93 (1H, dd, *J* = 8.0, 6.2 Hz, 3-H), 8.04 (2H, d, *J* = 8.9 Hz, Ph 2,6-H_2), 9.13 (1H, d, *J* = 7.5 Hz, 4-H), 9.25 (1H, d, *J* = 6.3 Hz, 2-H), 12.77 (1H, br, NH); ^{13}C NMR δ 44.0 (NMe), 57.0 (OMe), 127.0 (Ph 2,6-C2), 132.0 (Ph 3,5-C_2), 148.0 (4-C); MS *m*/*z* 267.1136 $(M)^+$ ($C_{16}H_{15}N_2O_2$ requires 267.1134).

4.1.20. 1-Methyl-5-oxo-7-(4-trifluoromethylphenyl)-1,6-naphthyridin-1-ium iodide (12d)

Compound **9d** (30 mg, 0.21 mmol) was stirred with MeI (90 mg, 0.63 mmol) in dry DMF (5 mL) for 72 h. The mixture was poured into EtOAc (3 mL). The solid was collected, washed (EtOAc) and dried to give **12d** (40 mg, 90%) as a yellow solid. mp 298–299 °C; ^{1}H NMR δ 4.45 (3H, s, Me), 7.31 (1H, s, 8-H), 7.59 (1H, m, 3-H), 7.99 (2H, d, J = 8.8 Hz, Ph 2,6-H$_2$), 8.17 (2H, d, J = 6.4 Hz, Ph 3,5-H$_2$), 9.15 (1H, d, J = 6.4 Hz, 4-H), 9.30 (1H, d, J = 4.4 Hz, 2-H), 13.00 (1H, br, NH); ^{13}C NMR δ 45.34 (Me), 95.52 (8-C), 122.44 (3-C), 123.64 (4a-C), 124.94 (q, J = 271.0 Hz, CF$_3$), 125.81 (q, J = 3.6 Hz, Ph 3,5-C$_2$), 129.21 (Ph 2,6-C$_2$), 131.16 (q, J = 34.0 Hz, Ph 4-C), 136.15 (7-C), 144.30 (3-C), 147.81 (8a-C), 149.89 (Ph 1-C), 151.41 (2-C), 160.46 (1-C); ^{19}F NMR ((CD$_3$)$_2$SO) δ −61.35 (s, CF$_3$); MS m/z 305.0895 (M)$^+$ ($C_{16}H_{12}F_3N_2O$ requires 305.0896).

4.1.21. 7-(4-Chlorophenyl)-1-methyl-5-oxo-1,6-naphthyridin-1-ium iodide (12e)

MeI (226 mg, 1.6 mmol) was stirred with **9e** (78 mg, 0.3 mmol) in dry DMF (5 mL) for 4 d. The evaporation residue was washed (EtOAc, petroleum ether) and dried to give **12e** (20 mg, 16%) as a yellow solid: mp >300 °C (decomp.); ^{1}H NMR δ 4.42 (3H, s, Me), 7.26 (1H, s, 8-H), 7.84 (1H, dd, J = 6.5, 5.8 Hz, 3-H), 7.99 (2H, d, J = 8.3 Hz, Ph 2,6-H$_2$), 8.31 (2H, d, J = 8.2 Hz, Ph 3,5-H$_2$), 9.07 (1H, d, J = 6.5 Hz, 4-H), 9.18 (1H, d, J = 5.8 Hz, 2-H), 12.53 (1H, br, NH); ^{13}C NMR δ 48.56 (Me), 90.0 (8-C), 118.0 (3-C), 127.0 (Ph 2,6-C$_2$), 130.0 (Ph 3,5-C$_2$), 151.0 (2-C); MS m/z 273.0626 (M)$^+$ ($C_{15}H_{12}{}^{37}ClN_2O$ requires 273.0608); 271.0634 (M)$^+$ ($C_{15}H_{12}{}^{35}ClN_2O$ requires 271.0638).

4.1.22. 7-(4-Bromophenyl)-1-methyl-5-oxo-1,6-naphthyridin-1-ium iodide (12f)

Compound **9f** was treated with MeI, as for the synthesis of **12a**, to give **12f** (52%) as a yellow solid: mp 292–294 °C, ^{1}H NMR δ 4.48 (3H, s, Me), 7.29 (1H, s, 8-H), 7.89 (2H, d, J = 8.7 Hz, Ph 2,6-H$_2$), 7.97 (1H, m, 3-H), 7.98 (2H, d, J = 8.7 Hz, Ph 3,5-H$_2$), 9.18 (1H, d, J = 7.9 Hz, 4-H), 9.30 (1H, d, J = 5.8 Hz, 2-H), 12.94 (1H, s, NH); ^{13}C NMR δ 45.98 (Me), 95.00 (8-C), 122.40 (3-C), 123.00 (4a-C), 126.00 (Ph 4-C), 130.12 (Ph 2,6-C$_2$), 130.50 (Ph 1-C), 131.98 (Ph 3,5-C$_2$), 132.55 (3-C), 144.50 (4-C), 148.50 (8a-C), 152.50 (2-C), 155.5 (1-C); MS m/z 317.0108 (M)$^+$ ($C_{15}H_{12}^{81}BrN_2NaO$ requires 317.0221), 315.0128 (M)$^+$ ($C_{15}H_{12}^{79}BrN_2NaO$ requires 315.0110).

4.1.23. 7-(4-Aminophenyl)-1-methyl-5-oxo-1,6-naphthyridin-1-ium iodide (12g)

MeI (144 mg, 1.02 mmol) was stirred with **9g** (42 mg, 0.19 mmol) in dry DMF (5 mL) for 4 d at room temperature, then at 80 °C for 2 h. The mixture was poured into EtOAc (25 mL) and petroleum ether (25 mL). The solid was collected, washed (petroleum ether) and dried to give **12g** (28 mg, 41%) as a yellow solid: mp >300 °C (decomp.); IR ν_{max} 3431, 3212, 1666, 1593, 1565 cm^{-1}; ^{1}H NMR δ 4.40 (3H, s, Me), 6.89 (2H, d, J = 9.0 Hz, Ph 3,5-H$_2$), 7.08 (1H, s, 8-H), 7.78 (1H, dd, J = 7.8, 6.2 Hz, 3-H), 7.98 (2H, d, J = 9.1 Hz, Ph 2,6-H$_2$), 9.03 (1H, d, J = 7.8 Hz, 4-H), 9.14 (1H, d, J = 5.2 Hz, 2-H), 12.53 (1H, br, NH); ^{13}C NMR δ 44.73 (Me), 90.53 (8-C), 111.59 (Ph 3,5-C$_2$), 117.33 (Ph 1-C), 119.33 (3-C), 122.18 (4a-C), 129.25 (Ph 2,6-C$_2$), 143.61 (7-C), 148.21 (2-C), 150.70 (Ph 4-C), 151.69 (4-C), 152.58 (8a-C), 160.61 (5-C); MS m/z 252 (M)$^+$.

4.1.24. 7-Phenyl-1,6-naphthyridin-5-one 1-oxide (13a)

Urea·H$_2$O$_2$ complex (78 mg, 0.8 mmol) was added to **9a** (100 mg, 0.45 mmol) in DMF (15 mL). The mixture was cooled to 0 °C. (F$_3$CCO)$_2$O (189 mg, 0.9 mmol) was added dropwise. The mixture was stirred at 0 °C for 12 h. The precipitate was collected and washed (water, CHCl$_3$).The solvent was evaporated from the combined filtrate and washings. Chromatography (petroleum ether/EtOAc 1:7 → AcOH/petroleum ether/EtOAc 1:3:21) gave **13a** (51 mg, 50%) as a white solid; mp 293–295 °C; IR ν_{max} 3434, 1678, 1620, 1585, 1505, 1244 cm^{-1}; ^{1}H NMR δ 7.31 (s, 8-H), 7.41 (1H, dd, J = 8.0, 6.5 Hz, 3-H), 7.53 (3H, m, Ph 3,4,5-H$_3$), 7.83 (2H, d, J = 8.0 Hz, Ph 2,6-H$_2$), 7.99 (1H, dd, J = 8.0, 1.0 Hz, 4-H), 8.62 (1H, dd, J = 6.5, 1.0 Hz, 2-H); ^{13}C NMR δ 93.77 (8-C), 121.90 (4a-C), 123.19 (3-C), 123.31 (Ph 4-C), 127.15 (Ph 2,6-C$_2$), 128.95 (Ph 3,5-C$_2$), 130.29 (4-C), 137.31 (Ph 1-C), 140.80 (7-C), 162.11 (8a-C); ^{15}N NMR δ 252.76 (s, 1-N), 129.56 (s, 6-N).

4.1.25. 7-(4-Methylphenyl)-1,6-naphthyridin-5-one 1-oxide (13b)

Urea·H$_2$O$_2$ complex (60 mg, 0.6 mmol) was added to **9b** (75 mg, 0.3 mmol) in DMF (10 mL). The mixture was cooled to 0 °C. (F$_3$CCO)$_2$O (123 mg, 0.6 mmol) was added dropwise. The mixture was stirred at 0 °C for 12 h. The precipitate was collected and washed (water, Me$_2$CO). The solvent was evaporated from the filtrates. Recrystallisation (EtOAc/petroleum ether) gave **13b** (20 mg, 29%) as a white solid; mp 258–260 °C; IR ν_{max} 3450, 1654, 1581, 1284 cm^{-1}; ^{1}H NMR δ 2.38 (3H, s, Me), 6.90 (1H, s, 8-H), 7.32 (2H, d, J = 8.5 Hz, Ph 3,5-H$_2$), 7.47 (1H, dd, J = 8.0, 4.5 Hz, 3-H), 7.73 (2H, d, J = 8.5 Hz, Ph 2,6-H$_2$), 8.50 (1H, dd, J = 7.5, 1.0 Hz, 4-H), 8.62 (1H, dd, J = 4.5, 1.5 Hz, 2-H), 11.78 (1H, s, 6-H); ^{13}C NMR δ 20.84 (Me), 103.99 (8-C), 121.49 (3-C), 126.86 (Ph 2,6-C$_2$), 129.42 (Ph 3,5-C$_2$), 135.03 (4-C), 154.96 (2-C); MS m/z 253.0996 (M+H)$^+$, ($C_{15}H_{13}N_2O_2$ requires 253.0972).

4.1.26. 2-Phenylpyrido[2,3-*d*]pyrimidin-4-one (14a)

Compound **38** (101.5 mg, 0.5 mmol), **45a** (60 mg, 0.5 mmol), CuI (19.2 mg, 0.1 mmol) and Cs$_2$CO$_3$ (325 mg, 1.0 mmol) were stirred in DMF (5 mL) under Ar for 10 h, then at 80 °C for 3 h, before being cooled and filtered. The evaporation residue, in MeOH, was filtered. The evaporation residue, in EtOAc/CH$_2$Cl$_2$, was filtered. The filtrate was washed (aq EDTA). The aq layer was extracted with CH$_2$Cl$_2$ (6×) and with EtOAc (6×). Drying and evaporation gave **14a** (58 mg, 53%) as an off-white solid: mp 285–287 °C (lit.[48] mp 284–285 °C); ^{1}H NMR (CDCl$_3$) δ 7.45 (1H, dd, 7-H), 7.60 (5H, m, Ph-H$_5$), 8.41 (1H, dd, J = 7.5, 2.0 Hz, 8-H), 8.91 (1H, dd, J = 4.5, 2.0 Hz, 6-H), 11.10 (1H, br, NH); ^{13}C NMR (CDCl$_3$) δ 116.11 (8a-C), 121.48 (7C), 128.49 (Ph 3,5-C$_2$), 128.98 (3C), 129.10 (Ph 2,6-C$_2$), 132.53 (Ph 4-C), 134.06 (Ph 1-C), 140.16 (8-C), 155.49 (6-C), 159.26 (4a-C), 166.46 (1-C); MS m/z 242.0957 (M+H$_2$O+H)$^+$ ($C_{13}H_{12}N_3O_2$ requires 242.0923), 224.0835 (M+H)$^+$ ($C_{13}H_{10}N_3O$ requires 224.0818).

4.1.27. 2-(4-Methylphenyl)pyrido[2,3-*d*]pyrimidin-4-one (14b)

Compound **38** (101.5 mg, 0.5 mmol) was stirred with **45b** (67 mg, 0.5 mmol), CuI (19.2 mg, 0.1 mmol) and Cs$_2$CO$_3$ (325 mg, 1.0 mmol) in DMF (5 mL) at 80 °C for 3 h, then at 20 °C for 10 h under Ar. The mixture was filtered. The evaporation residue, in MeOH, was filtered. The evaporation residue, in sat. aq EDTA (20 mL), was kept at 2–8 °C for 30 min and the solid was collected. The solid was washed (petroleum ether, water) and was dried to give **14b** (41 mg, 35%) as a white powder: mp 249–251 °C; ^{1}H NMR δ 2.47 (3H, s, Me), 7.28 (1H, dd, J = 7.4, 5.1 Hz, 6-H), 7.48 (2H, d, J = 8.1 Hz, Ph 3,5-H$_2$), 7.94 (2H, d, J = 8.1 Hz, Ph 2,6-H$_2$), 8.44 (1H, d, J = 7.5 Hz, 5-H), 8.48 (1H, d, J = 4.7 Hz, 7-H), 11.09 (1H, br, NH); ^{13}C NMR δ 20.99 (Me), 119.07 (4a-C), 119.84 (6-C), 127.31 (Ph 3,5-C$_2$), 129.53 (Ph 1-C), 129.61 (Ph 2,6-C$_2$), 140.15 (5-C), 142.84 (Ph 4-C), 148.29 (7-C), 155.60 (8a-C), 159.67 (2-C), 166.78 (4-C); MS m/z 236.0836 (M - H)$^-$ ($C_{14}H_{10}N_3O$ requires 236.0824).

4.1.28. 2-(4-Trifluoromethylphenyl)pyrido[2,3-*d*]pyrimidin-4-one (14d)

Compound **38** (101.5 mg, 0.5 mmol) was stirred with **45d** (94 mg, 0.5 mmol), CuI (19.2 mg, 0.1 mmol) and Cs$_2$CO$_3$ (325 mg,

1.0 mmol) in DMF (5 mL) at 80 °C for 3 h, then at 20 °C for 10 h under Ar. The mixture was filtered. The evaporation residue, in MeOH, was filtered. The evaporation residue was treated with sat. aq EDTA (20 mL) at 2–8 °C for 30 min. The solid was collected, washed (petroleum ether, water) and dried to give **14d** (64 mg, 45%) as a white powder: mp 254–256 °C; ^{1}H NMR δ 7.29 (1H, dd, J = 6.8, 4.0 Hz, 6-H), 8.04 (2H, m, Ph 2,6-H$_2$), 8.22 (2H, m, Ph 3,5-H$_2$), 8.46 (1H, d, J = 7.0 Hz, 5-H), 8.54 (1H, d, J = 4.0 Hz, 7-H), 11.01 (1H, br, NH); ^{13}C NMR δ 119.0 (6-C), 126.10 (Ph 2,6-C$_2$), 128.22 (q J = 2.7 Hz, Ph 3,5-C$_2$), 140.10 (5-C), 149.5 (7-C); ^{19}F NMR δ −61.35 (CF$_3$); MS 292.0699 (M+H)$^+$ ($C_{14}H_9F_3N_3O$ requires 292.0692).

4.1.29. 2-(4-Chlorophenyl)pyrido[2,3-*d*]pyrimidin-4-one (14e)

Compound **38** (101.5 mg, 0.5 mmol) was stirred with **45e** (77 mg, 0.5 mmol), CuI (19.2 mg, 0.1 mmol) and Cs_2CO_3 (325 mg, 1.0 mmol) in DMF (5 mL) at 80 °C for 3 h, then at 20 °C for 10 h under Ar. The mixture was filtered. The evaporation residue, in MeOH, was filtered. The evaporation residue was treated with sat. aq EDTA (20 mL) at 2–8 °C for 30 min. The solid was collected and recrystallised (EtOAc/CH_2Cl_2) to give **14e** (10 mg, 8%) as a pale grey powder: mp 266–268 °C (lit.[49] mp 300.5 °C); ^{1}H NMR δ 7.25 (1H, m, 6-H), 7.73 (2H, d, J = 8.9 Hz, Ph 2,6-H$_2$), 8.06 (2H, d, J = 8.9 Hz, Ph 3,5-H$_2$), 8.42 (1H, m, 5-H), 8.45 (1H, m, 7-H), 11.10 (1H, br NH); ^{13}C NMR δ 119.30 (4a-C), 119.75 (6-C), 129.27 (Ph 3,5-C$_2$), 129.50 (Ph 2,6-C$_2$), 132.0 (Ph 1-C), 137.0 (Ph 4-C), 140.25 (5-C), 149.34 (7-C), 151.0 (8a-C), 156.99 (2-C), 160.15 (1-C); MS m/z 537.0621 (2M+Na)$^+$ ($C_{26}H_{16}{}^{35}Cl_2N_6NaO_2$ requires 537.0598), 282.0225 (M+Na)$^+$ ($C_{13}H_8{}^{37}ClN_3NaO$ requires 282.0224), 280.0257 (M+Na)$^+$ ($C_{13}H_8{}^{35}ClN_3NaO$ requires 280.0254).

4.1.30. 2-Phenylpyrido[3,4-*d*]pyrimidin-4-one (15a)

Compound **31** (101.5 mg, 0.50 mmol) was stirred with **45a** (60 mg, 0.50 mmol), CuI (19 mg, 0.10 mmol) and Cs_2CO_3 (325 mg, 1.0 mmol) in DMF (5 mL) at 80 °C for 3 h, then at 20 °C for 12 h under Ar. The evaporation residue, in MeOH, was filtered. The evaporation residue, in sat. aq EDTA (20 mL), was sonicated for 5 min. The solution was kept at 2–8 °C for 1 h. The solid was collected. The filtrate was extracted with EtOAc (4×). The combined extracts were dried and the solvent was evaporated. Washing (water, petroleum ether) and drying gave **15a** (14 mg, 13%) as a white powder: mp 228–230 °C (lit.[50] mp 266–267 °C); ^{1}H NMR δ 7.62 (3H, m, Ph 3,4,5-H$_3$), 8.02 (1H, d, J = 5.0 Hz, 5-H), 8.26 (2H, d, J = 6.9 Hz, Ph 2,6-H$_2$), 8.68 (1H, d, J = 5.0 Hz, 6-H), 9.16 (1H, s, 8-H), 10.40 (1H, br, NH); ^{13}C NMR δ 118.09 (5-C), 127.42 (Ph 1-C), 127.93 (Ph 2,6-C$_2$), 128.97 (Ph 3,5-C$_2$), 129. 04 (4a-C), 131.56 (8a-C), 132.99 (Ph 4-C), 145.44 (6-C), 150.70 (8-C), 155.07 (2-C), 162.13 (4-C); MS m/z 469.1398 (2M+Na)$^+$ ($C_{26}H_{18}N_6NaO_2$ requires 469.1384), 246.0638 (M+Na)$^+$ ($C_{13}H_9N_3NaO$ requires 246.0641).

4.1.31. 2-(4-Methylphenyl)pyrido[3,4-*d*]pyrimidin-4-one (15b)

Compound **31** (101.5 mg, 0.50 mmol) was stirred with **45b** (67 mg, 0.50 mmol), CuI (19 mg, 0.10 mmol) and Cs_2CO_3 (325 mg, 1.0 mmol) in DMF (5 mL) at 80 °C for 3 h, then at 20 °C for 12 h under Ar. The evaporation residue, in MeOH, was filtered. The evaporation residue, in sat. aq EDTA (20 mL), was extracted thrice with CH_2Cl_2. Drying and evaporation gave **15b** (10 mg, 9%) as a white powder: mp 214–216 °C, ^{1}H NMR δ 2.44 (3H, s, Me), 7.36 (2H, d, J = 8.1 Hz, Ph 3,5-H$_2$), 7.91 (1H, d, J = 5.2 Hz, 5-H), 8.24 (2H, d, J = 8.2 Hz, Ph 2,6-H$_2$), 8.52 (1H, d, J = 5.2 Hz, 6-H), 9.03 (1H, s, 7-H), 10.20 (1H, br, NH); ^{13}C NMR δ 20.97 (Me), 118.16 (5-C), 126.00 (4a-C), 127.47 (Ph 1-C), 127.47 (Ph 2,6-C$_2$), 128.85 (Ph 3,5-C$_2$), 129.45 (Ph 4-C), 144.00 (6-C), 149.00 (8a-C), 150.40 (8-C), 168.90 (2-C); MS m/z 236.0836 (M - H)$^-$ ($C_{14}H_{10}N_3O$ requires 236.0824).

4.1.32. 2-(4-Trifluoromethylphenyl)pyrido[3,4-*d*]pyrimidin-4-one (15d)

Compound **31** (101.5 mg, 0.50 mmol) was stirred with **45d** (94 mg, 0.5 mmol), CuI (19.2 mmol, 0.1 mmol) and Cs_2CO_3 (325 mg, 1.0 mmol) in DMF (5 mL) at 80 °C for 3 h, then at 20 °C for 10 h under Ar. The mixture was filtered. The evaporation residue was suspended in MeOH and the suspension was filtered. The evaporation residue, sat. aq EDTA (20 mL) was kept at 2–8 °C for 1 h and the solid was collected. The filtrate was extracted with EtOAc (6×). The combined extracts were dried and the solvent was evaporated. Washing (petroleum ether, water) and drying gave **15d** (34 mg, 24%) as an off-white powder: mp >300 °C (decomp.); ^{1}H NMR δ 7.87 (2H, d, J = 8.3 Hz, Ph 2,6-H$_2$), 8.12 (1H, m, 5-H), 8.43 (2H, d, J = 8.2 Hz, Ph 3,5-H$_2$), 8.71 (1H, br, 6-H), 9.23 (1H, br, 8-H), 13.10 (1H, br, NH); ^{13}C NMR δ 125.24 (q, J = 289.3 Hz, CF$_3$), 125.56 (q, J = 2.3 Hz, Ph 3,5-C$_2$), 128.91 (Ph 2,6-C$_2$), 131.5 (4a-C), 136.27 (q, J = 31.7 Hz, Ph 4-C), 138.0 (Ph 1-C), 147.0 (5-C), 149.0 (8-C), 154.0 (2-C), 155.5 (6-C), 158.0 (8a-C), 161.32 (4-C); MS m/z 314 (M+Na)$^+$, 292.0705 (M+H)$^+$ ($C_{14}H_9F_3N_3O$ requires 292.0692).

4.1.33. 2-Phenylpyrido[4,3-*d*]pyrimidin-4-one (16a)

Compound **37** (101.5 mg, 0.5 mmol) was stirred with **45a** (60 mg, 0.5 mmol), CuI (19.2 mg, 0.1 mmol) and Cs_2CO_3 (325 mg, 1.0 mmol) in DMF (5 mL) at 80 °C for 3 h, then at 50 °C for 10 h under Ar. The mixture was filtered. The evaporation residue was suspended in MeOH and filtered. The evaporation residue, in sat. aq EDTA (20 mL), was sonicated for 5 min. This mixture was extracted with EtOAc (6×). Drying and evaporation gave **16a** (50 mg, 44%) as an off-white powder: mp 273–275 °C (lit.[51] 284–286 °C), ^{1}H NMR δ 7.61 (1H, m, 8-H), 7.63 (3H, m, Ph 3,4,5-H$_3$), 8.28 (2H, d, J = 7.2 Hz, Ph 2,6-H$_2$), 8.83 (1H, br, 7-H), 9.33 (1H, br, 5-H), 12.80 (1H, br, NH); ^{13}C NMR δ 117.5 (4a-C), 120.0 (8-C), 124.0 (Ph 1-C), 128.26 (Ph 3,5-C$_2$), 128.64 (Ph 2,6-C$_2$), 132.0 (Ph 4-C), 135.0 (3-C), 149.0 (5-C), 152.0 (7-C), 156.0 (8a-C), 162.0 (4-C); MS m/z 469.1396 (2M+Na)$^+$ ($C_{26}H_{18}N_6NaO_2$ requires 469.1389), 246.0644 (M+Na)$^+$ ($C_{13}H_9N_3NaO$ requires 246.0643), 224.0828 (M+H)$^+$ ($C_{13}H_{10}N_3O$ requires 224.0818).

4.1.34. 2-(4-Methylphenyl)pyrido[4,3-*d*]pyrimidin-4-one (16b)

Compound **37** (101.5 mg, 0.5 mmol) was stirred with **45b** (67 mg, 0.5 mmol), CuI (19.2 mmol, 0.1 mmol) and Cs_2CO_3 (325 mg, 1.0 mmol) in DMF (5 mL) at 80 °C for 12 h under Ar. The mixture was filtered. The evaporation residue was suspended in MeOH and filtered. The evaporation residue, in sat. aq EDTA (20 mL), was extracted with EtOAc (6×). Drying and evaporation gave **16b** (46 mg, 42%) as an off-white powder: mp 276–278 °C (lit.[51] 296–299 °C); ^{1}H NMR δ 2.45 (3H, s, Me), 7.29 (2H, d, J = 8.0 Hz, Ph 3,5-H$_2$), 7.64 (1H, d, J = 5.4 Hz, 5-H), 8.18 (2H, d, J = 8.2 Hz, Ph 2,6-H$_2$), 8.85 (1H, br, 6-H), 9.33 (1H, br, 8-H); ^{13}C NMR δ 21.0 (Me), 120.12 (8a-C), 120.39 (5-C), 128.17 (Ph 2,6-C$_2$), 129.21 (Ph 3,5-C$_2$), 129.82 (Ph 1-C), 131.48 (Ph 4-C), 142.27 (3-C), 149.45 (8-C), 157.37 (4a-C), 162.37 (1-C); MS m/z 238.0984 (M+H)$^+$ ($C_{14}H_{12}N_3O$ requires 238.0980).

4.1.35. 2-(4-Trifluoromethylphenyl)pyrido[4,3-*d*]pyrimidin-4-one (16d)

Compound **37** (101.5 mg, 0.5 mmol) was stirred with **45d** (94 mg, 0.5 mmol), CuI (19.2 mg, 0.1 mmol) and Cs_2CO_3 (325 mg, 1.0 mmol) in DMF (5 mL) at 80 °C for 3 h, then at 20 °C for 10 h under Ar. The mixture was filtered. The evaporation residue was suspended in MeOH. The suspension was filtered. The evaporation residue, in sat. aq EDTA, was kept at 2–8 °C for 1 h and the solid was collected. The filtrate was extracted with EtOAc (6×). The extracts were dried and the solvent was evaporated. Washing (petroleum ether, water) and drying gave **16d** (70 mg, 50%) as an off-white powder: mp 267–268 °C; ^{1}H NMR δ 7.71 (1H, d, J = 4.4 Hz,

8-H), 8.00 (2H, d, J = 8.3 Hz, Ph 2,6-H_2), 8.44 (2H, d, J = 8.2 Hz, Ph 3,5-H_2), 9.08 (1H, br, 7-H), 9.38 (1H, br, 5-H), 13.10 (1H, br, NH); ^{13}C NMR δ 122.04 (8a-C), 125.21 (8-C), 125.24 (Ph 2,6-C_2), 125.51 (q, J = 3.5 Hz, Ph 3,5-C_2), 129.17 (6-C), 131.76 (q, J = 340.6 Hz, CF_3), 134.0 (q, J = 27.5 Hz, Ph 4-C), 138.0 (Ph 1-C), 149.53 (5-C), 153.49 (4a-C), 156.32 (3-C), 162.0 (1-C); ^{19}F NMR δ −61.34 (s, CF_3); MS m/z 605 $(2M+Na)^+$, 314.0496 $(M+Na)^+$ ($C_{14}H_8F_3N_3NaO$ requires 314.0517), 292.0702 $(M+H)^+$ ($C_{14}H_9F_3N_3O$ requires 292.0698).

4.1.36. 2-(4-Chlorophenyl)pyrido[4,3-*d*]pyrimidin-4-one (16e)

Compound **37** (101.5 mg, 0.5 mmol) was stirred with **45e** (77.3 mg, 0.5 mmol), CuI (19.2 mmol, 0.1 mmol) and Cs_2CO_3 (325 mg, 1.0 mmol) in DMF (5 mL) at 80 °C for 12 h under Ar. The mixture was filtered. The evaporation residue, in MeOH, was filtered. The evaporation residue, in sat. aq EDTA (20 mL), was extracted with EtOAc (6×). The combined extracts were dried. The evaporation residue was washed (petroleum ether, water) and dried to give **16e** (17 mg, 13%) as a white powder: mp >300° (decomp.); ^{1}H NMR δ 7.66 (1H, d, J = 5.6 Hz, 5-H), 7.69 (2H, d, J = 8.6 Hz, Ph 2,6-H_2), 8.29 (2H, d, J = 8.6 Hz, Ph 3,5-H_2), 8.86 (1H, d, J = 5.5 Hz, 6-H), 9.34 (1H, br, 8-H), 13.05 (1H, br, NH); ^{13}C NMR δ 120.0 (5-C), 126.50 (8a-C), 128.78 (Ph 3,5-C_2), 130.09 (Ph 2,6-C_2), 131.02 (Ph 4-C), 137.15 (Ph 1-C), 146.0 (4-C), 153.52 (8-C), 155.99 (3-C), 162.0 (1-C); MS m/z 282.0269 $(M+Na)^+$ ($C_{13}H_8{}^{37}ClN_3NaO$ requires 282.0224), 280.0260 $(M+Na)^+$ ($C_{13}H_8{}^{35}ClN_3NaO$ requires 280.0254).

4.1.37. 1-Methyl-7-phenyl-1,2,3,4-tetrahydro-1,6-naphthyridin-5-one (17a)

BH_3·pyridine complex (60 μL) was added to **12a** (80 mg, 0.22 mmol) in HCO_2H (5 mL) at 0 °C was and the mixture was stirred for 5 d at 20 °C. Daily, additional BH_3·pyridine (10 μL) was added. Evaporation, trituration (MeOH; 10 mL) and recrystallisation (Pr^iOH) gave **17a** (21 mg, 20%) as a pale grey solid: mp 315–317 °C; IR ν_{max} 1655, 1620 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 1.89 (2H, m, 3-H_2), 2.51 (2H, m, 4-H_2), 3.11 (3H, s, Me), 3.36 (2H, m, 2-H_2), 6.30 (1H, s, 8-H), 7.43 (3H, m, Ph 3,4,5-H_3), 7.68 (2H, m, Ph 2,6-H_2); ^{13}C NMR ($CDCl_3$) δ 19.59 (4-C), 20.12 (Me), 50.78 (2-C), 96.80 (8-C), 100.22 (4a-C), 126.58 (Ph 2,6-C_2), 129.25 (Ph 3,5-C_2), 130.47 (4-C), 132.79 (Ph 1-C), 144.28 (7-C), 155.00 (8a-C), 158.96 (5-C); MS m/z 503 $(2M+Na)^+$, 263.1177 $(M+Na)$ ($C_{15}H_{16}N_2NaO$ requires 263.1160).

4.1.38. 7-(4-Methoxyphenyl)-1-methyl-1,2,3,4-tetrahydro-1,6-naphthyridin-5-one (17c)

BH_3·Py complex (0.10 mL) was added dropwise to **12c** (50 mg, 0.13 mmol) in HCO_2H (5 mL) at 0 °C and the mixture was stirred at 20 °C for 5 d. Daily, additional BH_3·Py (0.01 mL) was added. The evaporation residue, in AcOH (2 mL), was diluted with water (10 mL). The mixture was extracted (EtOAc, 3×). Evaporation and recrystallisation (water) gave **17c** (1.0 mg, 3%) as a white powder: mp 221–223 °C; ^{1}H NMR ($CDCl_3$) δ 1.93 (2H, qn, J = 6.2 Hz, 3-H_2), 2.61 (2H, t, J = 6.4 Hz, 4-H_2), 3.02 (3H, s, NMe), 3.28 (2H, t, J = 5.4 Hz, 2-H_2), 3.85 (3H, s, OMe), 5.98 (1H, s, 8-H), 6.96 (2H, d, J = 8.9 Hz, Ph 3,5-H_2), 7.47 (2H, d, J = 8.9 Hz, Ph 2,6-H_2); ^{13}C NMR ($CDCl_3$) δ 20.60 (4-C), 20.83 (3-C), 38.81 (NMe), 50.79 (2-C), 55.44 (OMe), 94.01 (8-C), 100.49 (4a-C), 114.62 (Ph 3,5-C_2), 126.95 (Ph 1-C), 127.18 (Ph 2,6-C_2), 142.51 (Ph 4-C), 153.33 (8a-C), 160.67 (7-C), 162.28 (5-C); MS m/z 563 $(2M+Na)^+$, 541 $(2M+H)^+$, 293.1259 $(M+Na)^+$ ($C_{16}H_{18}N_2NaO_2$ requires 293.1266), 271.1452 $(M+H)^+$ ($C_{16}H_{19}N_2O_2$ requires 271.1447).

4.1.39. 1-Methyl-7-(4-trifluoromethylphenyl)-1,2,3,4-tetrahydro-1,6-naphthyridin-5-one (17d)

BH_3·Py complex (0.10 mL) was added dropwise to **12d** (30 mg, 0.07 mmol) in HCOOH (5 mL) at 0 °C. The mixture was stirred for 10 d at 20 °C. Each day, additional BH_3·Py (0.01 mL) was added. The evaporation residue, in water (5 mL), was sonicated. The precipitate was collected and recrystallised (water) to give **17d** (5.0 mg, 24%) as a white powder: mp 287–288 °C; ^{1}H NMR ($CDCl_3$) δ 1.94 (2H, qn, J = 5.5 Hz, 3-H_2), 2.63 (2H, t, J = 6.0 Hz, 4-H_2), 3.31 (2H, t, J = 5.5 Hz, 2-H_2), 6.09 (1H, s, 8-H), 7.67 (2H, d, J = 8.5 Hz, Ph 2,6-H_2), 7.72 (2H, d, J = 8.5 Hz, Ph 3,5-H_2); ^{13}C NMR ($CDCl_3$) δ 20.61 (4-C), 20.94 (3-C), 50.01 (2-C), 96.10 (8-C), 101.76 (4a-C), 125.10 (q, J = 275 Hz, CF_3), 126.38 (m, Ph 2,3,5,6-C_4), 131.95 (q, J = 37.5 Hz, Ph 4-C), 140.02 (Ph 1-C), 141.80 (7-C), 149.10 (5-C), 157.22 (8a-C); ^{19}F NMR ($CDCl_3$) δ −62.79 (CF_3); MS m/z 639.2179 $(2M+Na)^+$ ($C_{32}H_{30}F_6N_4NaO_2$ requires 639.2171), 617.2372 $(2M+H)^+$ ($C_{32}H_{31}F_6N_4O_2$ requires 617.2351), 331.1061 $(M+Na)^+$ ($C_{16}H_{15}F_3N_2NaO$ requires 331.1034), 309.1239 $(M+H)^+$ ($C_{16}H_{16}F_3N_2O$ requires 309.1215).

4.1.40. 7-(4-Chlorophenyl)-1-methyl-1,2,3,4-tetrahydro-1,6-naphthyridin-5-one (17e)

Compound **12e** was treated with BH_3·Py complex, as for the synthesis of **17d**, to give **17e** (44%) as a white powder: mp 195–197 °C; ^{1}H NMR ($CDCl_3$) δ 1.95 (2H, qn, J = 6.4 Hz, 3-H_2), 2.60 (2H, t, J = 6.4 Hz, 4-H_2), 3.04 (3H, s, NMe), 3.30 (2H, t, J = 5.6 Hz, 2-H_2), 6.06 (1H, s, 8-H), 7.43 (2H, d, J = 8.6 Hz, Ph 3,5-H_2), 7.54 (2H, d, J = 8.7 Hz, Ph 2,6-H_2); ^{13}C NMR ($CDCl_3$) δ 20.62 (4-C), 20.83 (3-C), 38.76 (NMe), 50.43 (2-C), 95.28 (8-C), 101.97 (4a-C), 127.54 (Ph 2,6-C_2), 129.33 (Ph 3,5-C_2), 134.29 (Ph 1-C), 137.70 (Ph 4-C), 142.30 (7-C), 153.97 (8a-C), 157.50 (5-C); MS m/z 549.1819 $(2M+H)^+$ ($C_{30}H_{31}{}^{35}Cl_2N_4O_2$ requires 549.1824), 275.0941 $(M+H)^+$ ($C_{15}H_{16}{}^{35}ClN_2O$ requires 275.0946).

4.1.41. 7-(4-Bromophenyl)-1-methyl-1,2,3,4-tetrahydro-1,6-naphthyridin-5-one (17f)

Compound **12f** was treated with BH_3·Py complex, as for the synthesis of **17d**, to give **17f** (47%) as a white powder: mp 205–207 °C; ^{1}H NMR ($CDCl_3$) δ 1.95 (2H, qn, J = 6.4 Hz, 3-H_2), 2.61 (2H, t, J = 6.3 Hz, 4-H_2), 3.29 (3H, s, Me), 3.29 (2H, t, J = 5.5 Hz, 2-H_2), 6.02 (1H, s, 8-H), 7.42 (2H, d, J = 8.7 Hz, Ph 2,6-H_2), 7.59 (2H, d, J = 8.7 Hz, Ph 3,5-H_2); ^{13}C NMR ($CDCl_3$) δ 20.75 (4-C), 21.63 (3-C), 38.70 (NMe), 50.76 (2-C), 94.44 (8-C), 101.46 (4a-C),123.93 (Ph 4-C), 127.41 (Ph 2,6-C_2), 132.27 (Ph 3,5-C_2), 133.39 (Ph 1-C), 141.80 (7-C), 153.16 (8a-C), 162.35 (5-C); MS m/z 663/661/659 $(2M+Na)^+$, 641/639/637 $(2M+H)^+$, 343.0249 $(M+Na)^+$ ($C_{15}H_{15}{}^{81}BrN_2NaO$ requires 343.0246), 341.0264 $(M+Na)^+$ ($C_{15}H_{15}{}^{79}BrN_2NaO$ requires 341.0266), 321.0426 $(M+H)^+$ ($C_{15}H_{16}{}^{81}BrN_2O$ requires 321.0426), 319.0448 $(M+H)^+$ ($C_{15}H_{16}{}^{79}BrN_2O$ requires 319.0446).

4.1.42. 4-Oxo-2-phenyl-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidinium formate (18a)

Compound **47a** (50 mg, 0.16 mmol) was stirred with Pd/C (10%, 50 mg) in dry MeOH (10 mL) and HCO_2H (1.0 mL) for 10 h under Ar. Filtration (Celite®) and evaporation gave **18a** (32 mg, 89%) as an off-white powder: mp 244–245 °C (decomp.) (lit.[52] 214–216 °C for free base); ^{1}H NMR δ 2.46 (2H, m, 8-H_2), 3.20 (2H, m, 7-H_2), 3.79 (2H, s, 5-H_2), 7.59 (3H, m, Ph 3,4,5-H_3), 8.14 (2H, d, J = 7.2 Hz, Ph 2,6-H_2), 9.25 (2H, br, 3-NH, 7-NH); ^{13}C NMR δ 29.50 (8-C), 40.12 (5-C), 41.02 (7-C), 127.56 (Ph 3,5-C_2), 128.58 (Ph 2,6-C_2), 131.37 (Ph 4-C), 132.55 (Ph 1-C), 154.78 (4a-C), 158.03 (8a-C), 161.93 (2-C), 164.65 (4-C); MS m/z 477.2029 $(2M+Na)^+$ ($C_{26}H_{26}N_6NaO_2$ requires 477.2015), 250.0947 $(M+Na)^+$ ($C_{13}H_{13}N_3NaO$ requires 250.0956), 228.1169 $(M+H)^+$ ($C_{13}H_{14}N_3O$ requires 228.1137).

4.1.43. 2-(4-Methylphenyl)-4-oxo-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidinium formate (18b)

Compound **47b** was treated with Pd/C and HCO_2H in MeOH, as for the synthesis of **18a**, to give **18b** (86%) as an off-white powder:

mp 279–280 °C; ^{1}H NMR δ 2.42 (3H, s, Me), 2.45 (2H, m, 8-H_2), 2.73 (2H, m, 7-H_2), 3.33 (2H, s, 5-H_2), 7.36 (2H, d, J = 7.9 Hz, Ph 3,5-H_2), 8.03 (2H, d, J = 8.0 Hz, Ph 2,6-H_2), 8.31 (2H, br, 3-NH, 7-NH); ^{13}C NMR δ 20.94 (Me), 31.04 (7-C), 45.09 (8-C), 46.03 (5-C), 116.91 (4a-C), 127.45 (Ph 2,6-C_2), 129.16 (Ph 3,5-C_2), 129.50 (Ph 1-C), 141.41 (Ph 4-C), 154.08 (2-C), 158.07 (8a-C), 161.47 (4-C); MS m/z 242.1294 $(M+H)^+$ ($C_{14}H_{16}N_3O$ requires 242.1288).

4.1.44. 4-Oxo-2-(4-trifluoromethylphenyl)-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidinium formate (18d)

Compound **47d** (was treated with Pd/C and HCO_2H in MeOH, as for the synthesis of **18a**, to give **18d** (68%) as an off-white powder: mp 280–281 °C (decomp.); ^{1}H NMR δ 2.32 (2H, m, 8-H_2), 2.55 (2H, m, 7-H_2), 3.13 (2H, s, 5-H_2), 7.66 (2H, d, J = 8.5 Hz, Ph 3,5-H_2), 8.43 (2H, d, J = 8 Hz, Ph 2,6-H_2); ^{13}C NMR δ 31.52 (7-C), 46.07 (8-C), 53.41 (5-C), 114.42 (4a-C), 123.57 (q, J = 270.1 Hz, CF_3), 124.40 (q, J = 3.6 Hz, Ph 3,5-C_2), 127.77 (Ph 2,6-C_2), 128.41 (q, J = 31.1 Hz, Ph 4-C), 144.78 (Ph 1-C), 155.98 (8a-C), 156.99 (4-C), 159.29 (2-C); ^{19}F NMR δ −60.69 (s, CF_3); MS m/z 296.1016 $(M+H)^+$ ($C_{14}H_{13}F_3N_3O$ requires 296.1005).

4.1.45. 2-(4-Phenylethynylphenyl)-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidin-4-one hydrochloride (18j)

Compound **47j** (170 mg, 0.41 mmol) was stirred for 10 h with Pd/C (10%, 200 mg) in dry MeOH (10 mL) and HCOOH (1.0 mL). The mixture was filtered through Celite®. The evaporation residue was recrystallised (EtOH). The formate salt was treated with HCl in dioxane (4.0 M, 3.0 mL) and Et_2O (5 mL). The precipitate was collected and washed (petroleum ether) to give **18j** (44 mg, 34%) as a white crystalline powder: mp >260 °C (decomp.); ^{1}H NMR δ 2.92–2.95 (4H, m, 7,8-H_4), 4.02–4.04 (2H, m, 5-H_2), 7.24 (1H, m, Ph′ 4-H), 7.28 (2H, d, J = 7.3 Hz, Ph 3,5-H_2), 7.30–7.35 (4H, m, Ph′ 2,3,5,6-H_4), 7.41 (2H, d, J = 8.1 Hz, Ph 2,6-H_2), 8.26 (2H, br, +NH_2); ^{13}C NMR δ 36.52 (8-C), 36.86 (7-C), 41.88 (5-C), 66.32 (ethynyl 1-C), 92.21 (ethynyl 2-C), 128.21 (Ph′ 3,5-C_2), 128.33 (Ph 3,5-C_2), 128.37 (Ph 4-C), 128.53 (Ph′ 2,6-C_2), 128.68 (Ph′ 1-C), 128.77 (Ph 1-C), 128.86 (Ph 2,6-C_2), 131.49 (4a-C), 141.29 (8a-C), 141.82 (2-C), 167.31 (4-C); MS m/z 350.1263 $(M+Na)^+$ ($C_{21}H_{17}N_3NaO$ requires 350.1269).

4.1.46. 4-Oxo-2-(pyridin-4-yl)-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidinium formate (18l)

Compound **47l** was treated with Pd/C and HCO_2H in MeOH, as for the synthesis of **18a**, to give **18l** (80%) as an off-white powder: mp 242–244 °C (lit.[52] 244–246 °C); ^{1}H NMR δ 3.02 (2H, m, 8-H_2), 3.27 (2H, m, 7-H_2), 3.44 (2H, s, 5-H_2), 7.16 (2H, d, J = 8.2 Hz, Py 2,6-H_2), 7.53 (2H, d, J = 8.1 Hz, Py 3,5-H_2); ^{13}C NMR δ 31.14 (7-C), 45.32 (8-C), 50.61 (5-C), 116.89 (4a-C), 117.21 (Py 1-C), 125.53 (Py 2,6-C_2), 128.10 (Py 3,5-C_2), 156.85 (8a-C), 160.38 (4-C), 161.20 (2-C).

4.1.47. 3-Cyano-2-phenylethynylpyridine (25a)

Compound **24** (150 mg, 0.80 mmol) in THF (5 mL) was stirred with CuI (15.2 mg, 80 μmol) and $(Ph_3P)_2PdCl_2$ (28 mg, 40 μmol) in Pr^i_2NH (5 mL) under Ar at 45 °C for 30 min. Phenylethyne **21a** (163 mg, 1.6 mmol) was added. The mixture was stirred at 40 °C for 5 d. Evaporation and chromatography (petroleum ether/EtOAc 3:1) gave **25a** (80 mg, 50%) as a pale buff powder: mp 83–85 °C (lit.[53] mp 85–87 °C); ^{1}H NMR ($CDCl_3$) δ 7.33 (1H, dd, J = 8.0, 4.8 Hz, 5-H), 7.40 (3H, m, Ph 3,4,5-H_3), 7.68 (d, J = 7.6 Hz, Ph 2,6-H_2), 7.95 (1H, dd, J = 8.0, 2.0 Hz, 4-H), 8.77 (1H, dd, J = 4.8, 1.6 Hz, 6-H); ^{13}C NMR ($CDCl_3$) δ 85.62 (ethyne 2-C), 96.20 (ethyne 1-C), 112.82 (3-C), 115.91 (C≡N), 120.98 (5-C), 121.87 (Ph 1-C), 128.47 (Ph 3,5-C_2), 129.99 (Ph 4-C), 132.47 (Ph 2,6-C_2), 139.76 (4-C), 146.04 (2-C), 152.77 (6-C); MS m/z 227.0575 $(M+Na)^+$ ($C_{14}H_8N_2Na$ requires 227.0586), 205.0755 $(M+H)^+$ ($C_{14}H_9N_2$ requires 205.0766).

4.1.48. 3-Cyano-2-(4-methylphenylethynyl)pyridine (25b)

1-Ethynyl-4-methylbenzene **21b** was treated with **24**, CuI, Na ascorbate and $(Ph_3P)_2PdCl_2$ in Pr^i_2NH and THF, as for the synthesis of **25a**, except that the chromatographic eluent was petroleum ether/EtOAc (5:1 → 3:1), to give **25b** (80%) as a pale buff powder: mp 175–178 °C; ^{1}H NMR ($CDCl_3$) δ 2.40 (3H, s, Me), 7.21 (2H, J = 8.2 Hz, Ph 3,5-H_2), 7.33 (1H, dd, J = 8.0, 4.9 Hz, 5-H), 7.59 (2H, d, J = 8.1 Hz, Ph 2,6-H_2), 7.97 (1H, dd, J = 8.0, 1.8 Hz, 4-H), 8.78 (1H, dd, J = 4.9, 1.7 Hz, 6-H); ^{13}C NMR ($CDCl_3$) δ 21.70 (Me), 112.66 (2-C), 121.73 (5-C), 129.32 (Ph 3,5-C_2), 132.50 (Ph 2,6-C_2), 139.84 (4-C), 140.61 (Ph 4-C), 152.8 (6-C); MS m/z 241.0709 $(M+Na)^+$ ($C_{15}H_9N_2Na$ requires 241.0742), 219.0905 $(M+H)^+$ ($C_{15}H_{11}N_2$ requires 219.0922).

4.1.49. 3-Cyano-2-(4-methoxyphenylethynyl)pyridine (25c)

Compound **21c** was treated with **24**, CuI, Na ascorbate and $(Ph_3P)_2PdCl_2$ in Pr^i_2NH and THF, as for the synthesis of **25b**, to give **25c** (81%) as a pale buff powder: mp 118–120 °C; ^{1}H NMR ($CDCl_3$) δ 3.83 (3H, s, Me), 6.90 (2H, d, J = 8.9 Hz, Ph 3,5-H_2), 7.30 (1H, dd, J = 8.0, 4.9 Hz, 5-H), 7.62 (2H, d, J = 8.9 Hz, Ph 2,6-H_2), 7.93 (1H, dd, J = 8.2, 2.1 Hz, 4-H), 8.75 (1H, dd, J = 4.9, 1.7 Hz, 6-H); ^{13}C NMR δ 55.32 (Me), 85.01 (ethyne 2-C), 96.94 (ethyne 1-C), 112.94 (5-C), 114.22 (Ph 3,5-C_2), 116.09 (C≡N), 121.45 (3-C), 134.23 (Ph 2,6-C_2), 139.74 (4-C), 146.36 (6-C), 152.74 (2-C), 161.05 (Ph 4-C); MS m/z 257.0695 $(M+Na)^+$ ($C_{15}H_{10}N_2NaO$ requires 257.0690).

4.1.50. 3-Cyano-2-(4-trifluoromethylphenylethynyl)pyridine (25d)

Compound **21d** was treated with **24**, CuI, Na ascorbate and $(Ph_3P)_2PdCl_2$ in Pr^i_2NH and THF, as for the synthesis of **25b**, to give **25d** (55%) as a pale buff powder: mp 129–132 °C; ^{1}H NMR ($CDCl_3$) δ 7.40 (1H, dd, J = 7.9, 4.9 Hz, 5-H), 7.66 (2H, d, J = 8.0 Hz, Ph 3,5-H_2), 7.79 (2H, d, J = 8.0 Hz, Ph 2,6-H_2), 8.00 (1H, dd, J = 8.0, 1.8 Hz, 4-H), 8.81 (1H, dd, J = 4.9, 1.8 Hz, 6-H); ^{13}C NMR ($CDCl_3$) δ 87.25 (ethyne 2-C), 94.03 (ethyne 1-C), 113.22 (3-C), 115.78 (C≡N), 122.50 (5-C), 123 (CF_3), 124.73 (Ph 1-C), 125.49 (q, J = 3.6 Hz, Ph 3,5-C_2), 131.8 (q, J = ca. 30 Hz, Ph 4-C), 132.75 (Ph 2,6-C_2), 139.87 (4-C), 145.48 (2-C), 152.95 (6-C); ^{19}F NMR ($CDCl_3$) δ −63.03 (s, CF_3); MS m/z 295.0428 $(M+Na)^+$ ($C_{15}H_7F_3N_2Na$ requires 295.0459), 273.0622 $(M+H)^+$ ($C_{15}H_8N_2F_3$ requires 273.0640).

4.1.51. 2-(4-Chlorophenylethynyl)-3-cyanopyridine (25e)

Compound **21e** was treated with **24**, CuI, Na ascorbate and $(Ph_3P)_2PdCl_2$ in Pr^i_2NH and THF, as for the synthesis of **25b**, to give **25e** (42%) as an amber powder: mp 84–86 °C; ^{1}H NMR ($CDCl_3$) δ 7.36 (1H, dd, J = 7.9, 4.8 Hz, 5-H), 7.37 (2H, d, J = 8.7 Hz, Ph 3,5-H_2), 7.61 (2H, d, J = 8.7 Hz, Ph 2,6-H_2), 7.98 (1H, dd, J = 7.9, 1.7 Hz, 4-H), 8.79 (1H, dd, J = 5.0, 1.7 Hz, 6-H); ^{13}C NMR ($CDCl_3$) δ 86.38 (ethyne 2-C), 94.80 (ethyne 1-C), 112.87 (3-C), 115.82 (C≡N), 119.43 (Ph 1-C), 122.31 (5-C), 128.92 (Ph 3,5-C_2), 133.63 (Ph 2,6-C_2), 136.30 (Ph 4-C), 139.77 (4-C), 145.75 (2-C), 152.85 (6-C); MS m/z 239.0353 $(M+H)^+$ ($C_{14}H_8{}^{35}ClN_2H$ requires 239.0376).

4.1.52. 2-(4-Aminophenylethynyl)-3-cyanopyridine (25g)

Compound **21g** was treated with **24**, CuI, Na ascorbate and $(Ph_3P)_2PdCl_2$ in Pr^i_2NH and THF, as for the synthesis of **25b**, to give **25g** (364 mg, 87%), as a dark green powder: mp 128–130 °C; ^{1}H NMR ($CDCl_3$) δ 3.96 (2H, br, NH_2), 6.64 (2H, d, J = 9.1 Hz, Ph 3,5-H_2), 7.30 (1H, dd, J = 8.5, 1.8 Hz, 5-H), 7.49 (2H, d, J = 6.6 Hz, Ph 2,6-H_2), 7.92 (1H, dd, J = 7.9, 1.7 Hz, 4-H), 8.73 (1H, dd, J = 4.9, 1.7 Hz, 6-H); ^{13}C NMR ($CDCl_3$) δ 82.87 (ethyne 2-C), 98.10 (ethyne 1-C), 106.48 (Ph 1-C), 113.68 (Ph 3,5-C_2), 116.51 (5-C), 121.94 (C≡N), 133.58 (Ph 2,6-C_2), 140.65 (3-C), 145.43 (4-C), 150.12

(6-C), 151.16 (Ph 4-C), 153.27 (2-C); MS *m*/*z* 242.0693 $(M+Na)^+$ ($C_{14}H_9N_3Na$ requires 242.0694), 220.0876 $(M+H)^+$ ($C_{14}H_{10}N_3$ requires 220.0875).

4.1.53. 3-Cyano-2-((4-phenylmethoxyphenyl)ethynyl)pyridine (25h)

Compound **21h** was treated with **24**, CuI, Na ascorbate and $(Ph_3P)_2PdCl_2$ in Pr^i_2NH and THF, as for the synthesis of **25a**, to give **25h** (29%) as an off-white powder: mp 129–132 °C; IR ν_{max} 2215, 2189, 1599, 1506, 1464 cm^{-1}; ^1H NMR ($CDCl_3$) δ 5.08 (2H, s, CH_2), 6.98 (2H, d, J = 9.5 Hz, Ph 3,5-H_2) 7.29 (1H, dd, J = 7.9, 4.9 Hz, 5-H), 7.31–7.44 (5H, m, Ph′-H_5), 7.62 (2H, d, J = 9.5 Hz, Ph 2,6-H_2), 7.93 (1H, dd, J = 1.7 Hz, 7.9 Hz, 4-H), 8.74 (1H, dd, J = 4.9, 1.7 Hz, 6-H); ^{13}C NMR ($CDCl_3$) δ 70.06 (CH_2), 85.08 (C≡N), 96.85 (ethyne 2-C), 112.41 (ethyne 1-C), 113.22 (3-C), 115.11 (Ph 2,6-C_2), 116.08 (Ph 1-C), 121.47 (5-C), 127.40 (Ph′ 3,5-C_2), 128.11 (Ph′ 4-C), 128.60 (Ph′ 2,6-C_2), 134.24 (Ph 3,5-C_2), 136.26 (Ph′ 1-C), 139.73 (4-C), 146.33 (2-C), 152.73 (6-C), 160.21 (Ph 4-C); MS *m*/*z* 333 $(M+Na)^+$, 311.1181 $(M+H)^+$ ($C_{21}H_{15}N_2O$ requires 311.1179).

4.1.54. 3-Cyano-2-(pyridin-4-ylethynyl)pyridine (25l)

CuI (19.2 mg, 0.10 mmol), $(Ph_3P)_4Pd$ (57.8 mg, 0.05 mmol), **24** (184 mg, 1.0 mmol), **21l** (103 mg, 1.0 mmol) and Na ascorbate (19.8 mg, 0.10 mmol) were placed in a flask, which was degassed and filled with Ar. Pr^i_2NH (10 mL) and DMF (10 mL) were added. The mixture was stirred at 40 °C for 10 h. Evaporation and chromatography (petroleum ether/EtOAc 3:1 → 1:1 → 1:3) gave **25l** (58 mg, 28%) as an ivory-coloured powder: mp 110–113 °C; ^1H NMR ($CDCl_3$) δ 7.42 (1H, dd, J = 7.6, 4.9 Hz, 5-H), 7.52 (2H, d, J = 4.4 Hz, Py′ 3,5-H_2), 8.01 (1H, dd, J = 7.6, 1.8 Hz, 4-H), 8.65 (2H, d, J = 4.7 Hz, Py′ 2,6-H_2), 8.82 (1H, dd, J = 4.9, 1.8 Hz, 6-H); ^{13}C NMR ($CDCl_3$) δ 88.71 (ethyne 2-C), 92.19 (ethyne 1-C), 113.46 (5-C), 115.54 (C≡N), 123.01 (3-C), 125.84 (Py′ 3,5-C_2), 129.03 (Py′ 4-C), 139.85 (4-C), 149.97 (Py′ 2,6-C_2), 150.55 (6-C), 153.91 (2-C); MS *m*/*z* 206.0699 $(M+H)^+$ ($C_{13}H_8N_3$ requires 206.0719).

4.1.55. 7-(4-Methylphenyl)pyrano[4,3-*b*]pyridin-5-one (26b)

Compound **38** (101.5 mg, 0.5 mmol) was boiled under reflux with **37b** (252 mg, 1.0 mmol) and Cs_2CO_3 (163 mg, 0.5 mmol) in MeCN (15 mL) for 10 h. The mixture was cooled and poured into water (10 mL) and the mixture was extracted with CH_2Cl_2 (3×). The combined extracts were dried. The evaporation residue was suspended in petroleum ether (50 mL). The mixture was sonicated and was filtered. The collected solid was washed (water, petroleum ether) to give **26b** (110 mg, 93%) as an off-white powder: mp 135–136 °C; ^1H NMR ($CDCl_3$) δ 2.41 (3H, s, Me), 7.16 (1H, s, 8-H), 7.28 (2H, d, J = 8.0 Hz, Ph 3,5-H_2), 7.38 (1H, dd, J = 7.9, 4.7 Hz, 3-H), 7.80 (2H, d, J = 8.3 Hz, Ph 2,6-H_2), 8.52 (1H, ddd, J = 7.9, 1.7, 0.6 Hz 4-H), 8.92 (1H, dd, J = 4.7, 1.8 Hz, 2-H); ^{13}C NMR ($CDCl_3$) δ 21.0 (Me), 102.92 (8-C), 117.0 (4a-C), 123.0 (3-C), 125.55 (Ph 2,6-C_2), 126.0 (Ph 1-C), 129.68 (Ph 3,5-C_2), 137.52 (4-C), 141.0 (4-C), 150.0 (7-C), 156.33 (5-C), 158.0 (2-C), 162.0 (8a-C); MS *m*/*z* 238.0850 $(M+Na)^+$ ($C_{15}H_{12}NNaO_2$ requires 238.0863).

4.1.56. 7-(4-Methoxyphenyl)pyrano[4,3-*b*]pyridin-5-one (26c)

Compound **38** (101.5 mg, was treated with **37c**[42] and Cs_2CO_3, as for the synthesis of **26b**, to give **26c** (mg, 22%) as an off-white powder: mp 169–171 °C (lit.[54] 177–178 °C); ^1H NMR ($CDCl_3$) δ 3.78 (3H, s, Me), 6.63 (1H, s, 8-H), 6.88 (2H, d, J = 9.0 Hz, Ph 3,5-H_2), 7.28 (1H, dd, J = 7.5, 4.0 Hz, 3-H), 7.76 (2H, d, J = 8.5 Hz, Ph 2,6-H_2), 8.41 (1H, d, J = 8.0 Hz, 4-H), 8.81 (1H, m, 2-H); ^{13}C NMR ($CDCl_3$) δ 50.43 (Me), 91.46 (Ph 1-C), 101.86 (8-C), 113.77 (4a-C), 116.51 (Ph 3,5-C_2), 122.43 (3-C), 129.49 (Ph 2,6-C_2), 137.67 (4-C), 156.13 (2-C), 157.46 (7-C), 161.77 (8a-C), 163.66 (5-C); MS (ESI in MeOH, which converts **13c** to methyl 2-(2-(4-methoxyphenyl)-2-oxoethyl)pyridine-3-carboxylate) *m*/*z* 286.1075 $(M+H)^+$ ($C_{16}H_{16}NO_4$ requires 286.1082).

4.1.57. 7-(4-Chlorophenyl)pyrano[4,3-*b*]pyridin-5-one (26e)

Compound **38** (101.5 mg, 0.5 mmol) was boiled under reflux with **37e** (146.5 mg, 0.5 mmol) and Cs_2CO_3 (163 mg, 0.5 mmol) in MeCN (15 mL) for 12 h. The mixture was cooled and poured into water. The mixture was extracted (EtOAc, 3×). The combined extracts were washed (brine) and dried. The evaporation residue was washed (petroleum ether) to give **26e** (80 mg, 63%) as an off-white powder: mp 188–190 °C; ^1H NMR ($CDCl_3$) δ 7.13 (1H, s, 8-H), 7.36 (1H, dd, J = 8.0, 5.0 Hz, 3-H), 7.40 (2H, d, J = 8.5 Hz, Ph 3,5-H_2), 7.78 (2H, d, J = 8.5 Hz, Ph 2,6-H_2), 8.47 (1H, d, J = 8.0 Hz, 4-H), 8.87 (1H, d, J = 4.5 Hz, 2-H); ^{13}C NMR ($CDCl_3$) δ 117.02 (8a-C), 123.02 (3-C), 126.91 (Ph 3,5-C_2), 129.34 (Ph 2,6-C_2), 129.83 (8-C), 136.98 (Ph 4-C), 137.62 (4-C), 154.87 (7-C), 156.19 (4a-C), 156.49 (2-C), 161.78 (5-C); MS (ESI in MeOH, which converts **26e** into methyl 2-(2-(4-chlorophenyl)-2-oxoethyl)pyridine-3-carboxylate) *m*/*z* 292.0582 $(M+H)^+$ ($C_{15}H_{13}{}^{37}ClNO_3$ requires 292.0554), 290.0590 $(M+H)^+$ ($C_{15}H_{13}{}^{35}ClNO_3$ requires 290.0578).

4.1.58. 7-(4-Bromophenyl)pyrano[4,3-*b*]pyridin-5-one (26f)

Compound **38** was treated with **37f** and Cs_2CO_3, as for the synthesis of **26e**, to give **26f** (93%) as an off-white powder: mp 170–171 °C; ^1H NMR ($CDCl_3$) δ 7.25 (1H, s, 8-H), 7.47 (1H, dd, J = 8.0, 5.0 Hz, 3-H), 7.66 (2H, d, J = 8.5 Hz, Ph 3,5-H_2), 7.81 (2H, d, J = 8.5 Hz, Ph 2,6-H_2), 8.57 (1H, d, J = 8.0 Hz, 4-H), 8.97 (1H, d, J = 4.5 Hz, 2-H); ^{13}C NMR ($CDCl_3$) δ 104.01 (8-C), 117.01 (4a-C), 123.12 (3-C), 125.36 (Ph 4-C), 127.09 (Ph 2,6-C_2), 130.28 (Ph 1-C), 132.30 (Ph 3,5-C_2), 137.65 (4-C), 154.85 (8a-C), 156.27 (3-C), 156.49 (2-C), 161.78 (5-C); MS (ESI in MeOH, which converts **26f** into methyl 2-(2-(4-bromophenyl)-2-oxoethyl)pyridine-3-carboxylate) *m*/*z* 336.0074 $(M+H)^+$ ($C_{15}H_{13}{}^{81}BrNO_3$ requires 336.0054), 334.0063 $(M+H)^+$ ($C_{15}H_{13}{}^{79}BrNO_3$ requires 334.0073).

4.1.59. 3-Cyano-4-(phenylethynyl)pyridine (28a)

4-Bromo-3-cyanopyridine **27** (92 mg, 0.5 mmol) in THF (5 mL) was added to CuI (9.6 mg, 50 μmol), $(Ph_3P)_4Pd$ (29 mg, 25 μmol) and Na ascorbate (9.9 mg, 50 μmol) in Et_3N (5 mL) under Ar. The mixture was stirred at 40 °C for 30 min. Phenylethyne **21a** (76.5 mg, 0.75 mmol) was added and the mixture was stirred at 40 °C for 10 h. Evaporation and chromatography (petroleum ether/EtOAc 3:1) gave **28a** (80 mg, 78%) as an off-white powder: mp 74–75 °C (lit.[53] 85–87 °C); IR ν_{max} 2222, 2150, 1582, 1495 (C=N) cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.39–7.43 (3H, m, Ph 3,4,5-H_3), 7.45 (1H, d, J = 5.2 Hz, 5-H), 7.63 (2H, d, J = 8.2 Hz, Ph 2,6-H_2), 8.75 (1H, d, J = 5.2 Hz, 6-H), 8.87 (1H, s, 2-H); ^{13}C NMR ($CDCl_3$) δ 83.50 (ethyne 2-C), 101.29 (ethyne 1-C), 111.74 (4-C), 115.54 (C≡N), 120.76 (Ph 1-C), 125.06 (5-C), 128.59 (Ph 3,5-C_2), 130.28 (Ph 4-C), 132.38 (Ph 2,6-C_2), 134.87 (3-C), 152.40 (2-C), 152.68 (6-C); MS *m*/*z* 227.0569 $(M+Na)^+$ ($C_{14}H_8N_2Na$ requires 227.0585).

4.1.60. 3-Cyano-4-(4-methoxyphenylethynyl)pyridine (28c)

Compound **27** was treated with CuI, $(Ph_3P)_4Pd$, Na ascorbate and **21c** in Et_3N and THF, as for the synthesis of **28a**, except that the chromatographic eluent was petroleum ether/EtOAc (3:1 → 2:1), to give **28c** (68%) as a yellow powder: mp 101–102 °C; IR ν_{max} 2218, 2184, 1608, 1581, 1515 cm^{-1}; ^1H NMR ($CDCl_3$) δ 3.84 (3H, s, Me), 6.90 (2H, d, J = 9.0 Hz, Ph 3,5-H_2), 7.43 (1H, d, J = 5.3 Hz, 5-H), 7.57 (2H, d, J = 9.0 Hz, Ph 2,6-H_2), 8,71 (1H, d, J = 8.0, 6-H), 8.84 (1H, s, 2-H); ^{13}C NMR ($CDCl_3$) δ 55.34 (CH_3), 82.96 (ethyne 1-C), 102.12 (ethyne 2-C), 112.80 (3-C), 114.07 (C≡N), 114.30 (Ph 3,5-C_2), 115.66 (Ph 1-C), 124.72 (5-C), 134.18 (Ph 2,6-C_2), 135.31 (4-C), 152.23 (6-C), 152.63 (2-C), 161.25 (Ph 4-C); MS *m*/*z* 257.0695 $(M+Na)^+$ ($C_{15}H_{10}N_2NaO$ requires 257.0691).

4.1.61. 3-Cyano-4-(4-trifluoromethylphenylethynyl)pyridine (28d)

Compound **27** was treated with CuI, $(Ph_3P)_4Pd$, Na ascorbate and **21d** in Et_3N and THF, as for the synthesis of **28c**, to give **28d** (44%) as a yellow powder: mp 66–68 °C; IR ν_{max} 2232, 1612, 1568, 1173 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.49 (1H, d, J = 5.2 Hz, 5-H), 7.74 (2H, d, J = 8.2 Hz, Ph 3,5-H_2), 8.58 (1H, d, J = 5.4 Hz, 6-H), 8.80 (2H, d, J = 8.0 Hz, Ph 3,5-H_2), 8.91 (1H, s, 2-H); ^{13}C NMR ($CDCl_3$) δ 85.10 (ethyne 2-C), 101.20 (ethyne 1-C), 116.20 (3-C), 125.15 (5-C), 125.57 (Ph 2,6-C_2), 132.63 (Ph 3,5-C_2), 133.00 (4-C), 152.59 (6-C); ^{19}F NMR ($CDCl_3$) δ −63.07 (CF_3); MS m/z 273.0636 $(M+H)^+$ ($C_{15}H_8F_3N_2$ requires 273.0634).

4.1.62. 4-(4-Chlorophenylethynyl)-3-cyanopyridine (28e)

Compound **27** was treated with CuI, $(Ph_3P)_4Pd$, Na ascorbate and **21e** in Et_3N and THF, as for the synthesis of **28c**, to give **28e** (59%) as a yellow powder: mp 79–81 °C; IR ν_{max} 2232, 1569, 724 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.37 (2H, d, J = 8.4 Hz, Ph 2,6-H_2), 7.47 (1H, d, J = 5.2 Hz, 5-H), 7.55 (2H, d, J = 8.4 Hz, Ph 3,5-H_2), 8.75 (1H, d, J = 5.2 Hz, 6-H), 8.87 (1H, s, 2-H); ^{13}C NMR ($CDCl_3$) δ 99.55 (1-ethyne), 111.70 (3-C), 114.97 (C≡N), 119.28 (Ph 1-C), 124.96 (2-C), 129.05 (Ph 2,6-C_2), 133.57 (Ph 3,5-C_2), 137.80 (Ph 4-C), 152.45 (5-C), 152.70 (6-C); MS m/z 263.0156 $(M+Na)^+$ ($C_{14}H_7{}^{37}ClN_2Na$ requires 263.0166), 261.0183 $(M+Na)^+$ ($C_{14}H_7{}^{35}ClN_2Na$ requires 261.0196).

4.1.63. 4-(4-Aminophenylethynyl)-4-cyanopyridine (28g)

Compound **27** was treated with CuI, $(Ph_3P)_4Pd$, Na ascorbate and **21g** in Et_3N and THF, as for the synthesis of **28c**, to give **28g** (35%) as an orange powder: mp 155–159 °C; IR ν_{max} 3447, 2218, 2184, 1631, 1603, 1579 cm^{-1}; ^1H NMR ($CDCl_3$) δ 4.01 (2H, br, NH_2), 6.63 (2H, d, J = 8.7 Hz, Ph 3,5-H_2), 7.40 (1H, d, J = 4.5 Hz, 5-H), 7.44 (2H, d, J = 8.7 Hz, Ph 2,6-H_2), 8.67 (1H, d, J = 5.1 Hz, 6-H), 8.81 (1H, s, 2-H); ^{13}C NMR ($CDCl_3$) δ 82.71 (ethyne 1-C), 103.5 (ethyne 2-C), 109.71 (Ph 1-C), 111.22 (3-C), 114.56 (Ph 3,5-C_2), 124.49 (5-C), 134.23 (Ph 2,6-C_2), 136.10 (4-C), 148.55 (Ph 4-C), 152.09 (Ph 4-C), 152.61 (2-C); MS m/z 242.0681 $(M+Na)^+$ ($C_{14}H_9N_3Na$ requires 242.0694).

4.1.64. 2-Methyl-8-oxo-6-phenyl-2,7-naphthyridin-2-ium iodide (29a)

MeI (228 mg, 1.5 mmol) was stirred with **11a** (55 mg, 0.24 mmol) in dry DMF (5 mL) for 72 h. The mixture was poured into EtOAc (2 mL), followed by petroleum ether (20 mL). The solid was collected, washed (petroleum ether) and dried to give **29a** (10 mg, 11%) as a yellow solid: mp 280–283 °C (decomp.); ^1H NMR δ 4.41 (3H, s, Me), 7.28 (1H, s, 4-H), 7.64–7.69 (3H, m, Ph 3,4,5-H_3), 7.95 (2H, d, J = 8.2 Hz, Ph 2,6-H_2), 8.20 (1H, d, J = 6.7 Hz, 5-H), 8.82 (1H, d, J = 6.4 Hz, 6-H), 9.71 (1H, s, 8-H), 12.75 (1H, br, NH); ^{13}C NMR δ 35.77 (Me), 101.16 (4-C), 120.70 (8a-C), 123.33 (5-C), 127.75 (Ph 2,6-C_2), 129.12 (Ph 3,5-C_2), 131.61 (Ph 4-C), 132.12 (Ph 1-C), 143.76 (3-C), 147.25 (6-C), 147.47 (8-C), 152.06 (1-C), 160.83 (4a-C); MS m/z 237.1038 $(M+H)^+$ ($C_{15}H_{13}N_2O$ requires 237.1022).

4.1.65. 4-Cyano-3-(phenylethynyl)pyridine (35a)

Compound **34** (91.5 mg, 0.5 mmol) was stirred with $(PPh_3)_4Pd$ (30 mg, 25 μmol), CuI (9.6 mg, 50 μmol) and Na ascorbate (9.9 mg, 50 μmol) in THF (5 mL) and $Pr^i{}_2NH$ (5 mL) at 40 °C under Ar for 30 min. PhC≡CH **21a** (102 mg, 1.0 mmol) was added and the mixture was stirred at 40 °C under Ar for 16 h. Cooling, evaporation and chromatography (petroleum ether/EtOAc 4:1 → 1:3) gave **35a** (55 mg, 50%) as a pale buff powder: mp 46–48 °C (lit.[53] 49–53 °C); IR ν_{max} 2222, 2150, 1582 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.40 (3H, m, Ph 3,4,5-H_3), 7.53 (1H, d, J = 4.9 Hz, 5-H), 7.62 (2H, d, J = 9.6 Hz, Ph 2,6-H_2), 8.68 (1H, d, J = 4.9 Hz, 6-H), 8.92 (1H, s, 2-H); ^{13}C NMR ($CDCl_3$) δ 82.54 (1-C), 99.49 (2-C), 115.57 (C≡N), 121.28 (Ph 1-C), 122.35 (3-C), 124.95 (5-C), 128.36 (4-C), 128.52 (Ph 3,5-C_2), 129.78 (Ph 4-C), 132.09 (Ph 2,6-C_2), 148.40 (6-C), 152.77 (2-C); MS m/z 227.0590 $(M+Na)^+$ ($C_{14}H_8N_2Na$ requires 227.0580).

4.1.66. 4-Cyano-3-(4-methylphenylethynyl)pyridine (35b)

1-Ethynyl-4-methylbenzene **21b** was treated with **34**, CuI, $(PPh_3)_4Pd$ and Na ascorbate in THF and $Pr^i{}_2NH$, as for the synthesis of **35a**, except that the chromatographic eluent was petroleum ether/EtOAc (4:1 → 3:2) to give **35b** (62%) as a pale buff powder: mp 94–96 °C; ^1H NMR ($CDCl_3$) δ 2.39 (3H, s, Me), 7.20 (2H, d, J = 8.4 Hz, Ph 3,5-H_2), 7.51 (3H, m, 5-H, Ph 2,6-H_2), 8.65 (1H, d, J = 5.0 Hz, 6-H), 8.90 (1H, s, 2-H); ^{13}C NMR ($CDCl_3$) δ 21.62 (Me), 100.01 (ethynyl 1-C), 113.20 (C≡N), 114.90 (ethynyl 2-C), 119.50 (Ph 1-C), 123.50 (4-C), 124.90 (5-C), 129.15 (Ph 4-C), 129.31 (Ph 3,5-C_2), 132.03 (Ph 2,6-C_2), 140.50 (3-C), 148.15 (6-C), 152.02 (2-C); MS m/z 219.0910 $(M+H)^+$ ($C_{15}H_{11}N_2$ requires 219.0922).

4.1.67. 4-Cyano-3-(4-methoxyphenylethynyl)pyridine (35c)

Compound **21c** was treated with **34**, $(PPh_3)_4Pd$, CuI and Na ascorbate in THF and $Pr^i{}_2NH$, as for the synthesis of **35b**, to give **35c** (72%) as a pale buff powder: mp 96–97 °C; ^1H NMR ($CDCl_3$) δ 3.85 (3H, s, Me), 6.91 (2H, d, J = 8.9 Hz, Ph 3,5-H_2), 7.55 (1H, d, J = 5.0 Hz, 5-H), 7.56 (2H, d, J = 8.9 Hz, Ph 2,6-H_2), 8.64 (1H, d, J = 5.0 Hz, 6-H), 8.88 (1H, s, 2-H); ^{13}C NMR ($CDCl_3$) δ 55.35 (Me), 81.72 (ethynyl 1-C), 99.99 (ethynyl 2-C), 113.28 (Ph 1-C), 114.22 (Ph 3,5-C_2), 115.44 (C≡N), 121.92 (4-C), 122.93 (3-C), 124.87 (5-C), 133.77 (Ph 2,6-C_2), 147.92 (6-C), 152.57 (2-C), 160.81 (Ph 4-C); MS m/z 235.0854 $(M+H)^+$ ($C_{15}H_{10}N_2O$ requires 235.0827).

4.1.68. 4-Cyano-3-(4-trifluoromethylphenylethynyl)pyridine (35d)

Compound **21d** was treated with **34**, $(PPh_3)_4Pd$, CuI and Na ascorbate in THF and $Pr^i{}_2NH$, as for the synthesis of **35b**, to give **35d** (48%) as a pale orange powder: mp 39–40 °C; ^1H NMR ($CDCl_3$) δ 7.57 (1H, d, J = 5.0 Hz, 5-H), 7.67 (2H, d, J = 8.0 Hz, Ph 3,5-H_2), 7.74 (2H, d, J = 8.0 Hz, Ph 2,6-H_2), 8.73 (1H, d, J = 5.0 Hz, 6-H), 8.95 (1H, s, 2-H); ^{13}C NMR ($CDCl_3$) δ 84.49 (ethynyl 2-C), 97.52 (ethynyl 1-C), 115.15 (C≡N), 121.76 (3-C), 122.21 (4-C), 123.29 (q, J = 298.3 Hz, CF_3), 125.05 (5-C), 125.55 (q, J = 3.6 Hz, Ph 3,5-C_2), 131.45 (q, J = 32.8 Hz, Ph 4-C), 132.39 (Ph 2,6-C_2), 149.13 (6-C), 152.89 (2-C); ^{19}F NMR ($CDCl_3$) δ −63.00 (s, CF_3); MS m/z 295.0451 $(M+Na)^+$ ($C_{15}H_7F_3N_2Na$ requires 295.0454).

4.1.69. 3-(4-Chlorophenylethynyl)-4-cyanopyridine (35e)

Compound **21e** was treated with **34**, $(PPh_3)_4Pd$, CuI and Na ascorbate in THF and $Pr^i{}_2NH$, as for the synthesis of **35b**, to give **35e** (41%) as a pale buff powder: mp 200–201 °C; ^1H NMR ($CDCl_3$) δ 7.38 (2H, d, J = 6.7 Hz, Ph 2,6-H_2), 7.53 (1H, m, 5-H), 7.56 (2H, d, J = 6.7 Hz, Ph 3,5-H_2), 8.69 (1H, d, J = 5.5 Hz, 6-H), 8.91 (1H, s, 2-H); ^{13}C NMR ($CDCl_3$) δ 92.02 (ethynyl 2-C), 98.70 (ethynyl 1-C), 113.94 (C≡N), 118.96 (Ph 4-C), 119.21 (3-C), 123.90 (4-C),124.55 (5-C), 133.28 (Ph 3,5-C_2), 133.64 (Ph 2,6-C_2), 135.71 (Ph 1-C), 148.66 (6-C), 152.72 (2-C); MS m/z 239.0391 $(M+H)^+$ ($C_{14}H_8{}^{35}ClN_2$ requires 239.0371).

4.1.70. 7-(4-Methoxyphenyl)-2-methyl-5-oxo-2,6-naphthyridin-2-ium iodide (36c)

MeI (228 mg, 1.6 mmol) was stirred with **10c** (35 mg, 0.13 mmol) in dry DMF (5 mL) for 36 h. The mixture was poured into EtOAc (3 mL). The solid was collected, washed (Me_2CO) and dried to give **36c** (26 mg, 52%) as a yellow solid: mp 263–265 °C, ^1H NMR δ 3.91 (3H, s, OMe), 4.51 (3H, br, NMe), 7.15 (1H, s, 8-H), 7.18 (2H, d, J = 8.9 Hz, Ph 3,5-H_2), 7.85 (2H, d, J = 8.9 Hz, Ph 2,6-H_2), 8.63 (1H, d, J = 6.1 Hz, 4-H), 8.80 (1H, d, J = 6.4 Hz, 3-H), 9.59 (1H, br, NH); ^{13}C NMR δ 48.65 (NMe), 55.51 (OMe), 98.47 (8-C), 114.51 (Ph 3,5-C_2), 124.64 (4-C), 124.71 (Ph 1-C), 128.85

(Ph 2,6-C_2), 131.93 (8a-C), 138.94 (3-C), 146.18 (7-C), 147.21 (1-C), 160.08 (4a-C), 161.24 (Ph 4-C), 162.31 (5-C); MS *m*/*z* 267.1127 $(M)^+$ ($C_{16}H_{15}N_2O_2$ requires 267.1128).

4.1.71. 3-(4-Methylphenyl)pyrano[3,4-*c*]pyridin-1-one (40b)

Compound **39** (101.5 mg, 0.5 mmol) was heated under reflux with **37b** (252 mg, 1.0 mmol) and Cs_2CO_3 (163 mg, 0.5 mmol) in MeCN (15 mL) for 2 d. The mixture was cooled, poured into water and extracted thrice with CH_2Cl_2. The combined extracts were dried. The evaporation residue was suspended in petroleum ether (50 mL) and sonicated. The solid was collected and washed (water, petroleum ether) to give **40b** (38 mg, 32%) as an off-white powder: mp 120–123 °C; ^{1}H NMR ($CDCl_3$) δ 2.45 (3H, s, Me), 6.90 (1H, s, 4-H), 7.31 (2H, d, *J* = 8.0 Hz, Ph 3,5-H_2), 7.33 (1H, d, *J* = 5.5 Hz, 5-H), 7.83 (2H, d, *J* = 8.0 Hz, Ph 2,6-H_2), 8.83 (1H, d, *J* = 5.5 Hz, 6-H), 9.49 (1H, s, 8-H); ^{13}C NMR ($CDCl_3$) δ 20.5 (Me), 100.0 (4-C), 118.0 (4a-C), 118.5 (5-C), 121.5 (Ph 1-C), 125.80 (Ph 2,6-C_2), 128.9 (Ph 4-C), 129.79 (Ph 3,5-C_2), 130.0 (3-C), 139.5 (8-C), 141.0 (8a-C), 154.0 (6-C), 159.5 (1-C); MS *m*/*z* 238.0851 $(M+H)^+$ ($C_{15}H_{12}NO_2$ requires 238.0863).

4.1.72. 3-(4-Methoxyphenyl)pyrano[3,4-*c*]pyridin-1-one (40c)

Compound **39** (101.5 mg, 0.5 mmol) was heated under reflux with **37c** (142 mg, 0.5 mmol) and Cs_2CO_3 (163 mg, 0.5 mmol) in MeCN (15 mL) for 2 d. The cooled mixture was poured into water and extracted CH_2Cl_2 (3×). The combined extracts were washed (brine) and dried. The evaporation residue was washed (petroleum ether) to give **40c** (16.3 mg, 13%) as an off-white powder: mp 185–186 °C; ^{1}H NMR ($CDCl_3$) δ 3.91 (H, s, Me), 6.76 (1H, s, 4-H), 6.96 (2H, d, *J* = 8.5 Hz, Ph 3,5-H_2), 7.33 (1H, d, *J* = 5.5 Hz, 5-H), 7.88 (2H, d, *J* = 8.5 Hz, Ph 2,6-H_2), 8.81 (1H, d, *J* = 5.5 Hz, 6-H), 9.47 (1H, s, 8-H); ^{13}C NMR ($CDCl_3$) δ 55.49 (Me), 91.51 (4-C), 109.0 (Ph 1-C), 114.48 (Ph 3,5-C_2), 119.0 (5-C), 127.56 (Ph 4-C), 128.23 (Ph 2,6-C_2), 131.38 (3-C), 141.0 (8a-C), 143.0 (6-C), 152.8 (8-C), 163.03 (1-C); MS (ESI in MeOH, which converts **40c** to methyl 4-(2-(4-methoxyphenyl)-2-oxoethyl)pyridine-3-carboxylate) *m*/*z* 286.1074 $(M+H)^+$ ($C_{16}H_{16}NO_4$ requires 286.1082).

4.1.73. 3-(4-Bromophenyl)pyrano[3,4-*c*]pyridin-1-one (40f)

Compound **39** (101.5 mg, 0.5 mmol) was boiled under reflux with **37f** (382 mg, 1.0 mmol) and Cs_2CO_3 (163 mg, 0.5 mmol) in MeCN (15 mL) for 3 d. The cooled mixture was poured into water and extracted EtOAc (3×). The combined extracts were washed (brine) and dried. The evaporation residue was washed (petroleum ether) to give **40f** (10 mg, 7%) as a pale buff gum: ^{1}H NMR ($CDCl_3$) δ 6.83 (1H, s, 4-H), 7.36 (2H, d, *J* = 8.5 Hz, Ph 3,5-H_2), 7.69 (2H, d, *J* = 8.5 Hz, Ph 2,6-H_2), 7.90 (1H, dd, *J* = 8.5, 4.5 Hz, 5-H), 8.73 (1H, d, *J* = 4 Hz, 6-H), 9.37 (1H, s, 8-H); ^{13}C NMR ($CDCl_3$) δ 92.50 (4-C), 118.50 (3-C), 119.03 (4a-C), 127.35 (Ph 3,5-C_2), 129.86 (Ph 1-C), 132.05 (Ph 2,6-C_2), 134.50 (Ph 4-C), 143.46 (8a-C), 152.24 (8-C), 153.91 (6-C), 165.04 (1-C); MS (ESI in MeOH, which converts **40f** into methyl 4-(2-(4-bromophenyl)-2-oxoethyl)pyridine-3-carboxylate) *m*/*z* 336.0011 $(M+H)^+$ ($C_{15}H_{13}{}^{81}BrNO_3$ requires 336.0054), 334.0066 $(M+H)^+$ ($C_{15}H_{13}{}^{79}BrNO_3$ requires 334.0073).

4.1.74. 3-Phenylpyrano[4,3-*c*]pyridin-1-one (41a)

Compound **31** (101.5 mg, 0.5 mmol) was stirred with **37a** (114 mg, 0.5 mmol), K_3PO_4 (212 mg, 1.0 mmol) and CuI (10 mg, 50 μmol) in dry DMF (7 mL) at 100 °C under Ar for 6 d. Water (7 mL) was added to the cooled mixture. The mixture was extracted EtOAc (3×). The combined extracts were dried. Evaporation and chromatography (petroleum ether: EtOAc 5:1 → 3:1) gave **41a** (7.6 mg, 7%) as a yellow powder: mp 171–173 °C (lit.[53] 166–168 °C); ^{1}H NMR ($CDCl_3$) δ 7.02 (1H, s, 4-H), 7.48 (3H, m, Ph 3,4,5-H_3), 7.89 (2H, m, Ph 2,6-H_2), 8.06 (1H, d, *J* = 5.2 Hz, 8-H), 8.76 (1H, d, *J* = 5.1 Hz, 7-H), 8.97 (1H, s, 5-H); ^{13}C NMR δ 98.52 (4-C), 118.0 (8-C), 125.42 (Ph 2,6-C_2), 127.00 (8a-C), 129.01 (Ph 3,5-C_2), 130.64 (Ph 4-C), 131.00 (Ph 1-C), 132.00 (3-C), 148.44 (7-C), 149.00 (5-C), 151.00 (1-C); MS *m*/*z* 224.0708 $(M+H)^+$ ($C_{14}H_{10}NO_2$ requires 224.0712).

4.1.75. 6-Benzyl-2-phenyl-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidin-4-one (47a)

Compound **46** (1.42 g, 5.0 mmol) was boiled under reflux with **45a** (600 mg, 5.0 mmol) and NaOMe (from Na (690 mg, 30 mmol)) in dry MeOH (30 mL) for 10 h, then cooled. The evaporation residue, in water, was sonicated for 5 min. The suspension was kept at 2–8 °C for 1 h. The solid was collected, washed (water, EtOAc) and dried to give **47a** (840 mg, 53%) as an off-white powder: mp 233–235 °C; ^{1}H NMR δ 2.70–2.74 (4H, m, 7,8-H_4), 3.24 (2H, s, 5-H_2), 3.69 (2H, s, $PhCH_2N$), 7.28–7.37 (5H, m, Ph(Bn) 2,3,4,5,6-H_5), 7.47–7.53 (3H, m, Ph 3,4,5-H_3), 8.08 (2H, d, *J* = 7.0 Hz, Ph 2,6-H_2); 12.50 (1H, br, NH); ^{13}C NMR δ 31.38 (8-C), 49.12 (7-C), 49.45 (5-C), 61.70 (Ph-CH_2-N), 117.0 (4a-C), 127.01 (Ph(Bn) 4-C), 127.46 (Ph 2,6-C_2), 128.24 (Ph 2,6-C_2), 128.43 (Ph(Bn) 3,5-H_2), 128.78 (Ph(Bn) 2,6-H_2), 138.31 (Ph 1-C), 139.0 (Ph(Bn) Ph 1-C), 145.0 (8a-C), 153.5 (4-C), 159.0 (2-C); MS *m*/*z* 318.1586 $(M+H)^+$ ($C_{20}H_{20}N_3O$ requires 318.1601).

4.1.76. 6-Benzyl-2-(4-methylphenyl)-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidin-4-one (47b)

Compound **46** was treated with **45b** and NaOMe, as for the synthesis of **47a**, to give **47b** (56%) as an off-white powder: mp 234–235 °C; ^{1}H NMR δ 2.37 (3H, s, Me), 2.69–2.73 (4H, m, 7,8-H_4), 3.24 (2H, s, 5-H_2), 3.69 (2H, s, $PhCH_2N$), 7.29–7.31 (3H, m, Ph 4-H, Ar 3,5-H_2), 7.34–7.37 (4H, m, Ph 2,3,5,6-H_4), 7.98 (2H, d, *J* = 8.0 Hz, Ar 2,6-H_2); 12.50 (1H, br, NH); ^{13}C NMR δ 21.0 (Me), 32.0 (8-C), 49.5 (7-C), 49.8 (5-C), 62.0 ($PhCH_2N$), 125.8 (4a-C), 126.0 (Ph 4-C), 127.41 (Ar 2,6-C_2), 128.24 (Ar 3,5-C_2), 128.79 (Ph 2,6-C_2), 129.08 (Ph 3,5-C_2), 132.0 (Ar 1-C), 140.0 (Ph 1-C), 142.0 (Ar 4-C), 158.0 (8a-C), 161.5 (4-C), 175.0 (2-C); MS *m*/*z* 685 $(2M+Na)^+$, 663.3494 $(2M+H)^+$ ($C_{42}H_{43}N_6O_2$ requires 663.3447), 354.1592 $(M+Na)^+$ ($C_{21}H_{21}N_3NaO$ requires 354.1582), 332.1747 $(M+H)^+$ ($C_{21}H_{22}N_3O$ requires 332.1757).

4.1.77. 6-Benzyl-2-(4-trifluoromethylphenyl)-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidin-4-one (47d)

Compound **46** was treated with **45d** and NaOMe, as for the synthesis of **47a**, to give **47d** (60%) as an off-white powder: mp 234–235 °C; ^{1}H NMR δ 2.66–2.73 (4H, m, 7,8-H_4), 3.24 (2H, s, 5-H_2), 3.70 (2H, s, $PhCH_2N$), 7.39–7.41 (5H, m, Ph 2,3,4,5,6-H_5), 7.77 (2H, d, *J* = 7.8 Hz, Ar 3,5-H_2), 8.44 (2H, d, *J* = 7.7 Hz, Ar 2,6-H_2), 12.50 (1H, br, NH); ^{13}C NMR δ 31.47 (8-C), 50.14 (7-C), 50.64 (5-C), 62.19 ($PhCH_2N$), 115.38 (4a-C), 123.36 (q, *J* = 265.0 Hz, CF_3), 124.63 (q, *J* = 3.6 Hz, Ar 2,6-C_2), 126.86 (Ph 4-C), 127.88 (Ph 3,5-C_2), 128.15 (Ar 2,6-C_2), 128.60 (q, *J* = 32 Hz, Ar 4-C),, 128.74 (Ph 2,6-C_2), 138.74 (Ph 1-C), 142.81 (Ar1-C), 156.72 (8a-C), 157.82 (2-C), 169.10 (4-C); ^{19}F NMR δ −60.77 (CF_3); MS *m*/*z* 408 $(M+Na)^+$, 386.1493 $(M+H)^+$ ($C_{21}H_{19}F_3N_3O$ requires 386.1475).

4.1.78. 6-Benzyl-2-(4-chlorophenyl)-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidin-4-one (47e)

Compound **46** was treated with **45e** and NaOMe, as for the synthesis of **47a**, to give **47e** (58%) as an off-white powder: mp 211–213 °C; ^{1}H NMR δ 2.75–2.78 (4H, m, 7,8-H_4), 3.29 (2H, s, 5-H_2), 3.74 (2H, s, $PhCH_2N$), 7.34–7.42 (5H, m, Ph 2,3,4,5,6-H_5), 7.61 (2H, d, *J* = 8.2 Hz, Ar 3,5-H_2), 8.15 (2H, d, *J* = 7.9 Hz, Ar 2,6-H_2); 12.65 (1H, br, NH); ^{13}C NMR δ 32.00 (8-C), 49.80 (7-C), 49.90 (5-C), 62.00 ($PhCH_2N$), 128.50 (Ar 3,5-C_2), 128.79 (m, Ph 2,3,4,5,6-C_5), 129.50 (8a-C), 129.80 (Ar 2,6-C_2), 139.00 (4a-C), 154.00 (1-C); MS *m*/*z* 350.1066 $(M+H)^+$ ($C_{20}H_{17}{}^{35}ClN_3O$ requires 350.1066).

4.1.79. 6-Benzyl-2-(4-bromophenyl)-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidin-4-one (47f)

Compound **46** was treated with **45f** and NaOMe, as for the synthesis of **47a**, to give **47f** (43%) as an off-white powder: mp 253–255 °C; ^{1}H NMR δ 2.70–2.74 (4H, m, 7,8-H_4), 3.24 (2H, s, 5-H_2), 3.69 (2H, s, PhCH_2N), 7.28–7.37 (5H, m, Ph 2,3,4,5,6-H_5), 7.71 (2H, d, J = 9.0 Hz, Ar 3,5-H_2), 8.05 (2H, d, J = 8.5 Hz, Ar 2,6-H_2); 12.60 (1H, br, NH); ^{13}C NMR δ 32.0 (8-C), 49.5 (7-C), 49.8 (5-C), 62.0 (PhCH_2N), 118.7 (4a-C), 127.03 (Ph 4-C), 128.24 (Ph 2,3,5,6-C_4), 128.78 (Ar 2,6-C_2), 129.52 (Ar 3,5-C_2), 131.49 (Ar 1-C), 139.0 (Ph 1-C), 156.0 (8a-C), 158.0 (8a-C), 163.0 (4-C), 163.5 (4-C); MS m/z 398.0665 $(M+H)^+$ ($C_{20}H_{19}{}^{81}BrN_3O$ requires 398.0688), 396.0686 $(M+H)^+$ ($C_{20}H_{19}{}^{79}BrN_3O$ requires 396.0706).

4.1.80. 6-Benzyl-2-(4-phenylethynylphenyl)-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidin-4(3*H*)-one (47j)

NaOMe (259 mg, 4.8 mmol) in dry MeOH (5 mL) was added to **46** (216 mg, 0.8 mmol) in dry MeOH (5 mL), followed by **45j** (177.5 mg, 0.8 mmol) in dry MeOH (5 mL). The mixture was stirred at reflux for 16 h. The evaporation residue was recrystallised (water) to give **47j** (57%) as an off-white powder: mp 66–67 °C; ^{1}H NMR δ 2.36–2.66 (2H, m, 8-H_2), 2.86–3.25 (2H, m, 7-H_2), 3.46–3.63 (2H, m, 5-H_2), 3.68 (2H, s, PhCH_2N), 7.24–7.38 (4H, m, Ph′-2,3,5,6-H_4), 7.45–7.52 (4H, m, Ph(Bn)-2,3,5,6-H_4), 7.60–7.69 (2H, m, Ph′-4H, Ph(Bn) 4-H), 7.80 (2H, d, J = 8.1 Hz, Ph 3,5-H_2), 7.96 (2H, d, J = 8.0 Hz, Ph 2,6-H_2); ^{13}C NMR δ 29.73 (8-C), 50.22 (7-C), 51.78 (5-C), 59.94 (Bn–CH_2–N), 87.91 (ethynyl 1-C), 93.27 (ethynyl 2-C), 110.95 (Ph 4-C), 118.44 (Ph′ 1-C), 121.39 (Bn Ph 1-C), 126.99 (Ph 1-C), 128.77 (Ph′ 2,3,5,6-H_4), 128.86 (Bn Ph 2,3,5,6-H_4), 129.53 (Ph′ 4-C), 131.62 (Bn Ph 4-C), 132.13 (Ph 3,5-C_2), 132.60 (Ph 2,6-C_2), 138.33 (8a-C), 165.54 (4-C), 169.11 (2-C); MS m/z 418.1983 $(M+H)^+$ ($C_{28}H_{24}N_3O$ requires 418.1919).

4.1.81. 6-Benzyl-2-(pyridin-4-yl)-5,6,7,8-tetrahydropyrido[4,3-*d*]pyrimidin-4-one (47l)

Compound **46** was treated with **45l** and NaOMe, as for the synthesis of **47a**, to give **47l** (1.27 g, 86%) as an off-white powder: mp 298–300 °C; ^{1}H NMR δ 2.63 (2H, m, 8-H_2), 2.71 (2H, m, 7-H_2), 3.23 (2H, s, 5-H_2), 3.69 (2H, s, PhCH_2N), 7.31–7.42 (5H, m, Ph 2,3,4,5,6-H_5), 8.16 (2H, d, J = 4.5 Hz, Py 3,5-H_2), 8.58 (2H, d, J = 4.5 Hz, Py 2,6-H_2), 12.50 (1H, br, NH); ^{13}C NMR δ 31.20 (8-C), 50.50 (7-C), 53.70 (5-C), 62.50 (PhCH_2N), 117.60 (8a-C), 121.55 (Py 3,5-C_2), 128.13 (Ph 4-C), 128.73 (Ph 2,3,5,6-C_4), 139.5 (Ph 1-C), 148.10 (Py 1-C), 149.80 (Py 2,6-C_2), 157.00 (4a-C), 160.10 (3-C); 182.60 (1-C); MS m/z 341.1374 $(M+Na)^+$ ($C_{19}H_{18}N_4NaO$ requires 341.1378), 319.1532 $(M+H)^+$ ($C_{19}H_{19}N_4O$ requires 319.1559).

4.2. Protein crystallography

Protein crystallography experiments were carried out using human TNKS-2. The catalytic domain was expressed, purified and crystallised as previously reported.[29] Inhibitors were soaked for 48 h into the crystals in a well solution (0.2M $LiSO_4$, 0.1 M Tris–HCl pH 8.5, 24–26% PEG 3350) supplemented with the compound (100 μM) and NaCl (250 mM). Before collection of data, the crystals were briefly soaked in a well solution supplemented with 20% glycerol and flash frozen in liquid N_2.

Data were collected at the Diamond Light Source on beamline I04-1. Diffraction data were processed and scaled with the XDS package.[55] The structures were solved using the Difference Fourier method, with the starting phases derived from the TNKS-2 structure (PDB code 3U9H). REFMAC5[56] was used for refinement and COOT[57] for manual building of the model. Statistics for collection of data and refinement are shown in the Supplementary data.

Data for structures of **12b** and **17a** have been deposited at the Protein Data Bank with PDB codes 4W5I and 4UX4, respectively.

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Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.bmc.2015.05.005.

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