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Structural Studies on Bioactive Compounds. Part 29: Palladium Catalysed Arylations and Alkynylations of Sterically Hindered Immunomodulatory 2-Amino-5-halo-4,6-(disubstituted)pyrimidines†

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Abstract—The immunological agent bropirimine **5** is a tetra-substituted pyrimidine with anticancer and interferon-inducing properties. Synthetic routes to novel 5-aryl analogues of bropirimine have been developed and their potential molecular recognition properties analysed by molecular modelling methods. Sterically challenged 2-amino-5-halo-6-phenylpyrimidin-4-ones (halo = Br or I) are poor substrates for palladium catalysed Suzuki cross-coupling reactions with benzeneboronic acid because the basic conditions of the reaction converts the amphoteric pyrimidinones to their unreactive enolic forms. Palladium-mediated reductive dehalogenation of the pyrimidinone substrates effectively competes with cross-coupling. 2-Amino-5-halo-4-methoxy-6-phenylpyrimidines can be converted to a range of 5-aryl derivatives with the 5-iodopyrimidines being the most efficient substrates. Hydrolysis of the 2-amino-5-aryl-4-methoxy-6-phenylpyrimidines affords the required pyrimidin-4-ones in high yields. Semi-empirical quantum mechanical calculations show how the nature of the 5-substituent influences the equilibrium between the 1*H*- and 3*H*-tautomeric forms, and the rotational freedom about the bond connecting the 6-phenyl group and the pyrimidine ring. Both of these factors may influence the biological properties of these compounds.

Introduction

Synthesis of the small molecule immunomodulatory agent 2-amino-5-bromo-6-phenylpyrimidin-4(3*H*)-one (bropirimine: **5**) was first reported in 1975.[1] The drug, which can induce endogenous liberation of interferons[2,3] and provoke cytokine release leading to enhanced natural killer cell activity,[4] has been developed primarily as an alternative to the immunotherapeutic agent BCG vaccine for the oral treatment of bladder cancer.[5] This type of agent might also have uses to reduce the frequency of relapses in patients with progressive multiple sclerosis.

An intriguing feature of bropirimine is exhibited in its X-ray crystal structure. Bropirimine crystallises from *N*-methylformamide as a triply hydrogen-bonded duplex (Fig. 1) in which one molecule of the 3*H*-tautomer forms three hydrogens bonds to another molecule of the 1*H*-tautomer (Fig. 1) in the manner of a Watson–Crick cytosine–guanine base pair. Indeed the lengths of the *H*-bonds are almost precisely those in a cytosine–guanine duplex in DNA.[6] Thus bropirimine and potentially other molecules which possess a 2-aminodiazin-4-one fragment, and which display a divergent range of biological properties, have the potential to recognise guanine or cytosine residues in single-stranded RNA or DNA by Watson–Crick bonding or base sequences in double-stranded DNA by Hoogsteen bonds. Possibly, the immunomodulatory properties of bropirimine are triggered by these encounters (discussed in ref 6). Even if this is not the case, the crystal structure highlights two characteristics of bropirimine, either or both of which may be implicated in its biological properties: firstly, its array of potential hydrogen bond donors and acceptors; and secondly, its ability to adopt one of two tautomeric forms in response to its molecular environment.

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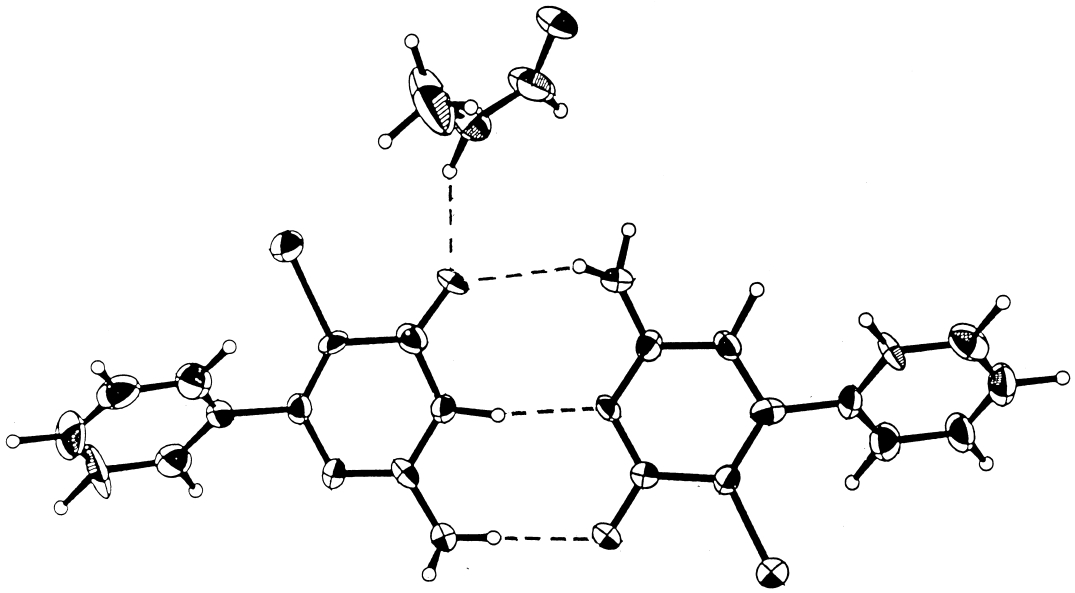


Figure 1. Hydrogen bonding between 2-amino-5-bromo-6-phenylpyrimidin-4(3*H*)-one and 2-amino-5-bromo-6-phenylpyrimidin-4(1*H*)-one in the crystal structure of the *N*-methylformamide solvate of bropirimine **5** (from ref 6). The 4(3*H*)-one tautomer is shown (left), the 4(1*H*)-one tautomer (right) and the molecule of *N*-methylformamide (above).

The presence of a halogen atom at C(5) is crucial for activity in this series of pyrimidinones.[2–4] However, it is difficult to provide an overall interpretation for the structure–activity investigations that have been performed on these compounds, since the results are dependent on the biological activity sought.[3]

From the crystal structure of bropirimine it is apparent that steric clashes between the bromine atoms and the phenyl groups in the duplex are ameliorated by in-plane bending so that both the Br-C(5)-C(6) and Ph-C(6)-C(5) angles exceed 120° and by twisting between the pyrimidine and phenyl ring-planes of 52 and 47° in the 3*H*- and 1*H*-tautomers, respectively (see Fig. 1).[6]

Replacement of the 5-bromo-6-phenyl fragment by a 5-aryl-6-phenyl structure would be expected to induce major perturbations in the disposition of substituents around the pyrimidine ring and modulate both the hydrogen-bonding potential of the molecules and the relative stability of the 1*H*- and 3*H*-tautomers. 2-Amino-5,6-diarylpyrimidin-4(3*H*)-ones have been little studied: previous syntheses have employed the traditional approach to 2-aminopyrimidinone synthesis of an initial condensation between guanidine carbonate and a 3-carbon synthon such as diphenylacrylonitrile[7] or diphenylcyclopropenone.[9] The Suzuki cross-coupling[9] procedure offers the potential of a more adaptable method to introduce a range of substituted phenyl residues into the 5-position of bropirimine. Although 5-halopyrimidines have been coupled with arylboronic acids under palladium-catalysed conditions, these earlier efforts have focused on neutral halopyrimidine substrates.[10] In contrast, 2-amino-5-halo-6-phenylpyrimidin-4-ones are amphoteric; the pK_a values corresponding to protonation at N(1) and deprotonation at N(3) of bropirimine **5** are 3.18 and 8.53, respectively.[11] Normally a requirement for efficient Suzuki coupling is that the halide component should be electron deficient. However, under the basic conditions normally employed in Suzuki couplings the halopyrimidinone substrate would be in an electron rich anionic state. To offset this potential disadvantage it is known that 5-halo substituents in pyrimidin-4-ones are labile. For example, debromination of bropirimine occurs readily in the presence of strong nucleophiles such as aqueous alkali and hydrazine hydrate[12] where nucleophilic attack at the electrophilic C(6) site presages displacement of the halogen atom at C(5); also, debromination occurs readily by thermal homolysis.[13] However, the aforementioned mechanisms are quite distinct from that of a Suzuki reaction which would require the in situ formation of an organopalladium (pyrimidinone-Pd-halide) intermediate.

Then there is the steric question. Approach of an arylboronic acid to the organopalladium intermediate would be impeded by the flanking carbonyl group at C(4) (or its enolate equivalent) and the bulky substituent at C(6). As the synthetic objectives of this work were to develop high-yielding cross-couplings from bropirimine and related pyrimidin-4-ones to yield their 5-aryl derivatives, this seemed to be a major impediment to a successful outcome that would demand exploration of a wide range of coupling conditions.

Chemistry

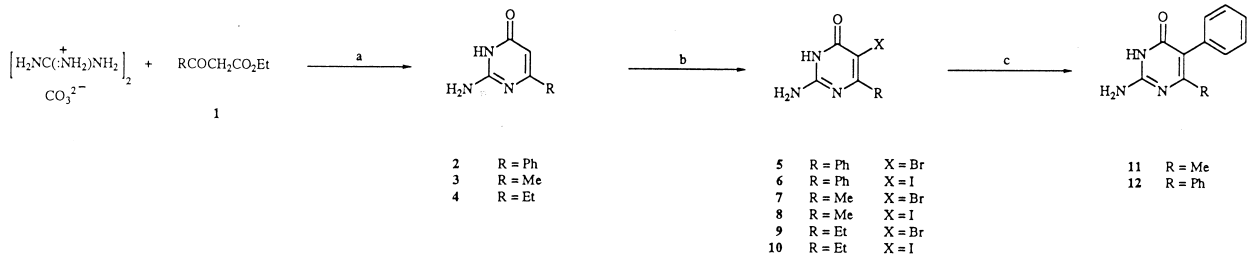
Arylation of 2-amino-5-halo-6-(substituted)pyrimidin-4-ones

The halopyrimidinones required for this work were synthesised in an efficient two-step process from guanidine carbonate and β-ketoesters (**1**: R = Ph, Me, Et). The pyrimidinones **2**–**4** formed in near quantitative

yields were either brominated (bromine in acetic acid) or iodinated (iodine in chloroform–aqueous sodium hydroxide) to afford a series of 5-bromo- and 5-iodo-pyrimidinones **5–10** (Scheme 1).

In order to minimise the steric impediment imposed by the 6-substituent initial Suzuki couplings employed the 5-bromo-6-methylpyrimidinone (**7**). This was reacted with benzeneboronic acid in degassed refluxing DMF in the presence of tetrakis(triphenylphosphine)palladium (0) [$Pd(PPh_3)_4$] (3.5 mol%) and aqueous sodium carbonate (3 mol equiv). The products were the desired 6-methyl-5-phenylpyrimidinone (**11**) (10%) and the debrominated methylpyrimidinone (**3**) (85%). Applying alternative 'standard' conditions used by Wang and Haseltine,[14] which employ toluene–ethanol as solvent and aqueous sodium carbonate (2 mol equiv) as base, only a trace of coupling (TLC) was observed and >90% starting material was recovered.

When bropirimine (**5**) was used as substrate in DMF the products were the required 5,6-diphenylpyrimidinone (**12**) (17%) and the debrominated product (**2**) (83%) (Table 1: Entry 1): use of triethylamine as base instead of sodium carbonate in DMF gave no coupled product (Table 1: Entry 2) whereas in toluene–ethanol a mixture comprising the coupled product **12** (9%), the debrominated pyrimidinone (**2**) (19%) and unreacted starting material (**5**) (72%) was formed (Table 1: Entry 3). Compound **12** was identical to an authentic sample prepared from diphenylcyclopropenone and guanidine carbonate.[8] Although disappointing these initial results showed, contrary to expectation, that the more sterically-challenged bromophenylpyrimidinone **5** was a marginally better substrate for coupling than the bromomethylpyrimidinone **7**; also use of toluene–ethanol was associated with less debromination. Based on this information a series of couplings was performed in toluene–ethanol in an effort to maximise the yield of



Scheme 1. Reagents: (a) EtOH reflux; (b) Br_2 in AcOH (X = Br), or I_2 in $CHCl_3$-aq. NaOH (X = I); (c) Suzuki cross-coupling (see Table 1).

Table 1. Products of cross-coupling reactions on 2-amino-5-halo-4,6-(disubstituted)pyrimidines: variations in reaction conditions[a]

Entry	Starting pyrimidine	Boronic acid	Solvent[b,g]	Catalyst	Base	Hours reflux	Products (% yield)[c]		
							Coupling	Dehalogenation	Unchanged
1	**5**	benzene	A	$Pd(PPh_3)_4$	Na_2CO_3	24	**12** (17)	**2** (83)	**5** (0)
2	**5**	benzene	A	$Pd(PPh_3)_4$	NEt_3	24	**12** (0)	**2** (29)	**5** (48)
3	**5**	benzene	B	$Pd(PPh_3)_4$	Na_2CO_3	24	**12** (9)	**2** (19)	**5** (72)
4	**5**	benzene	B	$Pd(PPh_3)_4$	$NaHCO_3$	24	**12** (4)	**2** (18)	**5** (56)
5	**6**	benzene	B	$Pd(PPh_3)_4$	Na_2CO_3	24	**12** (1)	**2** (30)	**6** (38)
6	**5**	benzene	B	$Pd(PPh_3)_4$	Na_2CO_3	24	**12** (15)	**2** (31)	**5** (34)
7	**5**	benzene	B	$Pd(PPh_3)_4$	Na_2CO_3	24	**12** (16)	**2** (25)	**5** (37)
8	**5**	benzene	B	$Pd(PPh_3)_2$	Na_2CO_3	18	**12** (10)	**2** (23)	**5** (31)
9	**5**	benzene	B	$Pd(PPh_3)_4$	Na_2CO_3	76	**12** (21)	**2** (46)	**5** (20)
10	**14**	benzene	B	$Pd(PPh_3)_4$	Na_2CO_3	18	**18** (75)[d]	[e]	[f]
11	**14**	2,4-dichlorobenzene	B	$Pd(PPh_3)_4$	Na_2CO_3	18	**19** (24)	**16** (11)	**14** (58)
12	**14**	2,4-dichlorobenzene	C	$Pd(dppf)(OAc)_2$	K_3PO_4	24	**19** (0)	**16** (24)	**14** (71)
13	**14**	2,4-dichlorobenzene	B	$Pd(dppf)(OAc)_2$	K_3PO_4	6	**19** (0)	**16** (29)	**14** (67)
14	**14**	2,4-dichlorobenzene	D	$Pd(OAc)_2$	Na_2CO_3	14	**19** (0)	**16** (13)	**14** (77)
15	**14**	3-thienyl	B	$Pd(PPh_3)_4$	Na_2CO_3	18	**20** (1)	**16** (89)	**14** (1)
16	**14**	3-thienyl	C	$Pd(dppf)(OAc)_2$	K_3PO_4	24	**20** (50)[d]	**16** (16)	**14** (24)
17	**14**	4-methoxybenzene	C	$Pd(dppf)(OAc)_2$	K_3PO_4	90	**21** (55)[d]	**16** (18)	**14** (20)
18	**17**	2,4-dichlorobenzene	C	$Pd(dppf)(OAc)_2$	K_3PO_4	5	**19** (0)	**16** (22)	**17** (67)
19	**17**	4-methoxybenzene	C	$Pd(dppf)(OAc)_2$	K_3PO_4	24	**21** (72)[d]	**16** (6)	**17** (0)
20	**17**	4-chlorobenzene	C	$Pd(dppf)(OAc)_2$	K_3PO_4	16	**22** (78)[d]	**16** (4)	**17** (0)
21	**17**	3-nitrobenzene	C	$Pd(dppf)(OAc)_2$	K_3PO_4	16	**23** (75)[d]	**16** (3)	**17** (0)

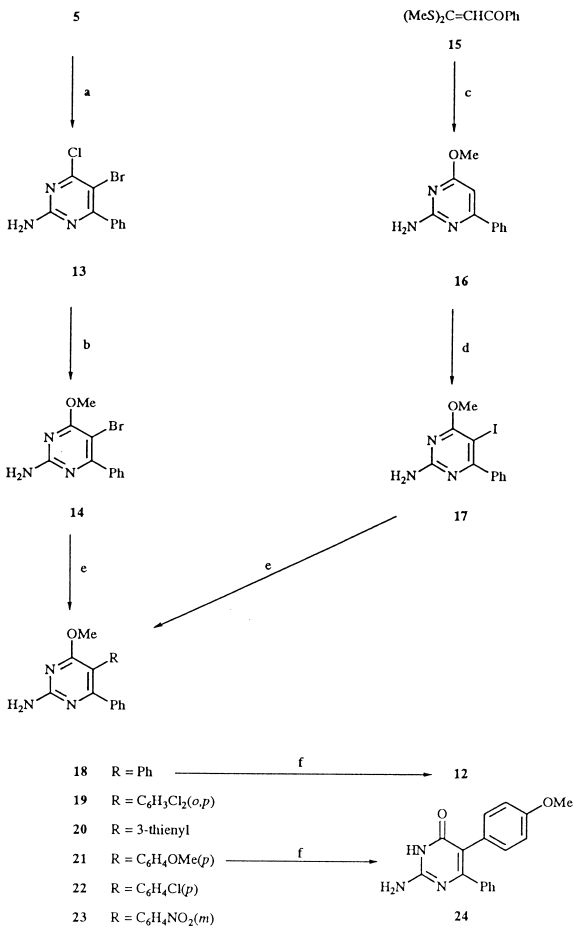
[a]See Experimental for details of syntheses.
[b]Including water; see Experimental for details.
[c]Relative yields determined by ^1H NMR analysis of partially purified (column chromatography) reaction mixtures.
[d]Isolated by crystallisation from the reaction mixture.
[e]De-halogenated product **2** detected (TLC) in the reaction mixture.
[f]Starting material **14** detected (TLC) in the reaction mixture.
[g]Solvents: A, dimethylformamide; B, toluene–ethanol; C, 1,2-dimethoxyethane; D, 95% ethanol.

the required diphenylpyrimidinone **12**. Product mixtures were purified by column chromatography and yields of pyrimidine products were analysed by ^{1}H NMR. Results are summarised in Table 1. Replacement of the sodium carbonate base with sodium bicarbonate led to a reduced yield of the coupled product (Table 1: Entry 4). Surprisingly, use of the 5-iodo-6-phenylpyrimidinone (**6**) as the halopyrimidinone component gave only a trace (TLC) of the coupled product **12** (Table 1: Entry 5). In all cases (above) unreacted starting material was recovered and considerable dehalogenation to **2** occurred in a process that must be mediated by palladium since **5** is stable in the reaction medium (refluxing toluene–ethanol containing sodium carbonate). Batch variation in the tetrakis(triphenylphosphine)-palladium(0) catalyst had a minimal bearing on the outcome of the couplings (Table 1; Entries 3, 6, 7); in situ generation of the presumed catalytic species, bis(triphenylphosphine)palladium(0) [$Pd(PPh_3)_2$], formed by reaction of palladium diacetate and triphenylphosphine[15] did not improve the outcome (Table 1: Entry 8). The highest yield of **12** (21%) was formed in toluene–ethanol over 76 h with periodic additions of more benzeneboronic acid, sodium carbonate and catalyst but the ratio of coupling: debromination still remained only 1:2.2 (Table 1: Entry 9). Presumably the poor yield of coupled product, even under the latter non-catalytic conditions, reflects the small proportion of neutral halopyrimidine substrate available for oxidative addition to the palladium(0) species involved in the catalytic cycle.[16]

Arylation of 2-amino-5-halo-4-methoxy-6-phenylpyrimidines

To explore the possibility that pre-derivatisation of the pyrimidinone moiety might facilitate more effective cross-coupling to 5-arylpyrimidines the 2-amino-5-halo-4-methoxy-6-phenylpyrimidines **14** and **17** were synthesised and subjected to Suzuki couplings (Scheme 2). Bropirimine **5** was first converted to the 4-chloropyrimidine **13** (67%) in refluxing phosphorus oxychloride. The product was accompanied by an insoluble polymeric material which was reconverted to bropirimine in boiling 1 M-hydrochloric acid. Possibly, this unidentified material was formed by condensation of the reactive chloro group of **13** with the 2-amino group [or N(1) or N(3) atoms] of another molecule. Methanolysis of **13** with sodium methoxide–methanol afforded the 5-bromo-4-methoxy-pyrimidine **14**. The corresponding 5-iodopyrimidine (**17**) was not available by this route since the 5-iodopyrimidinone (**6**) underwent quantitative de-iodination in phosphorus oxychloride. Instead, 3,3-bis(methylthio)-1-phenyl-2-propene-1-one (**15**) was cyclised to 2-amino-4-methoxy-6-phenylpyrimidine (**16**) with guanidine sulphate in sodium methoxide solution according to the method of Chauhan and Junjappa.[17] Iodination of **16** with *N*-iodosuccinimide in chloroform gave the required 5-iodopyrimidine (**17**).

Results of Suzuki couplings on halomethoxypyrimidines **14** and **17** are recorded in Table 1. Reaction between the 5-bromo-4-methoxypyrimidine (**14**) and benzeneboronic acid in toluene–ethanol with sodium carbonate base and tetrakis(triphenylphosphine)-palladium(0) catalyst



Scheme 2. Reagents: (a) $POCl_3$, reflux; (b) NaOMe, MeOH; (c) guanidinium sulphate, NaOMe, reflux; (d) *N*-iodosuccinimide, $CHCl_3$; (e) Suzuki reactions (see Table 1); (f) 10 M−HCl, MeOH, reflux.

afforded the 5,6-diphenyl-pyrimidine **18** in 75% yield (after crystallisation) (Table 1: Entry 10). But perversely, these conditions were not general and although 2,4-dichlorobenzeneboronic acid did yield the required 5-(2,4-dichlorophenyl)pyrimidine (**19**) in poor yield (24%), a pure sample was obtained only after repeated chromatographic fractionations to remove starting material **14** and debrominated product **16** (Table 1: Entry 11). The structure of **19**, which was presumably a mixture of enantiomeric (atropisomeric) pyrimidines non-interconvertible by restricted rotation about the pivotal pyrimidine–dichlorophenyl bond,[18] was confirmed by ^{1}H and ^{13}C NMR spectra and mass spectrometry.

Aqueous 1,2-dimethoxyethane (DME) has been recommended by Gronowitz as a substitute for toluene in troublesome couplings;[19] alternative bases including thallium (I) hydroxide and tripotassium phosphate have been reported to produce dramatic rate and yield enhancements in the synthesis of hindered biaryls.[20] Also the nature and quality of the palladium catalyst is often decisive: thus Thompson et al. have exploited the catalyst 1,1′-bis(diphenylphosphino)-ferrocenepalladium (II) acetate [$Pd(dppf)(OAc)_2$], prepared in situ, for cross-couplings of hindered halopyrazines and halopyr-

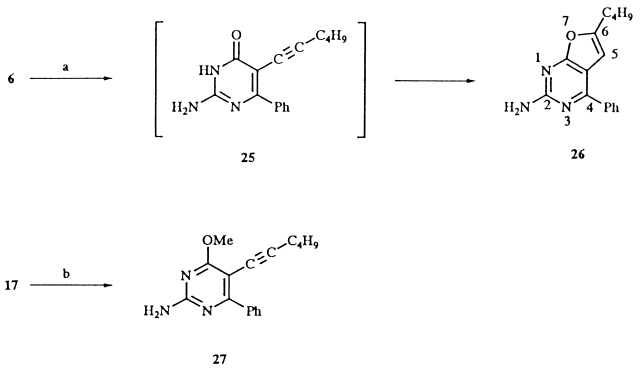
idines where tetrakis(triphenylphosphine)palladium(0) had failed;[21] and Campi et al. have shown that catalytic palladium acetate [$Pd(OAc)_2$] in degassed 95% ethanol containing sodium carbonate base can effect efficient cross-couplings of *ortho*-substituted benzeneboronic acids.[22]

Exploration of some of these variables to improve yields of the hindered dichlorophenylpyrimidine (**19**) met with no success (Table 1: Entries 12–14). However, coupling of **14** and 3-thienylboronic acid, which had given only a trace of product **20** with the toluene–ethanol–sodium carbonate–$Pd(PPh_3)_4$ combination, gave a 50% yield of **20** using DME and catalytic $Pd(dppf)(OAc)_2$ (Table 1: Entries 15, 16). Similarly the 5-(4-methoxyphenyl)-pyrimidine **21** was isolated (55%), together with debrominated by-product **16** and unreacted starting material using the latter conditions (Table 1: Entry 17). Reactions with the 5-iodo-4-methoxypyrimidine (**17**) and four boronic acids were attempted in DME-tripotassium phosphate-$Pd(dppf)(OAc)_2$. Again, 2,4-dichlorobenzeneboronic acid failed to give the desired product **19** (Table 1: Entry 18). However, reactions with 4-methoxy-, 4-chloro- and 3-nitrobenzeneboronic acids under these conditions furnished excellent yields of the 5-aryl-6-phenylpyrimidines **21–23**, respectively (Table 1: Entries 19–21): not only was cross-coupling more rapid between the 5-iodopyrimidine (**17**) and 4-methoxybenzene-boronic acid than with the 5-bromo-pyrimidine (**14**), but also selectivity for coupled (**21**) versus dehalogenated (**16**) product was enhanced from 3:1, starting with **14**, to 12:1 from **17**.

Hydrolysis of the 4-methoxy-5-phenylpyrimidine (**18**) in methanolic hydrochloric acid gave the pyrimidinone **12** (96%), identical to the sample originally prepared in poor yield directly from a Suzuki reaction on bropirimine. Similarly the 5-(4-methoxyphenyl)pyrimidinone (**24**) was formed (97%) by hydrolysis of the methoxy precursor **21**.

Alkynylation of 2-amino-5-iodo-6-phenylpyrimidines

Our preliminary results show that it is also possible to exploit Heck reactions to introduce alkynyl groups into sterically-hindered 2-amino-5-halo-4,6-(disubstituted)-pyrimidines. Interaction of the iodopyrimidinone **6** with 1-hexyne was conducted under standard Heck conditions [23] employing a mixture of triethylamine, DMF (to dissolve the pyrimidinone), and a mixed $Pd(PPh_3)_2Cl_2$-CuI catalyst at 50 °C. Identified products included starting iodopyrimidinone **6** (64%), deiodinated material **2** (11%) and a mixture of non-acidic pyrimidines which could not be separated completely. The major non-acidic product had spectroscopic characteristics consistent with a furo[2,3-*d*]pyrimidine structure **26**. Presumably the target 5-hexynylpyrimidinone (**25**) cyclised to the furopyrimidine in the reaction conditions (Scheme 3). Similar intramolecular Michael-type cyclisations have been reported in 5-alkynyluracils.[24] The proposed bicyclic structure was confirmed by a NOE experiment which showed the expected enhancements between H(5) and the α- and β-methylene protons of the



Scheme 3. Reagents: (a) NEt_3, DMF, 1-hexyne, $Pd(PPh_3)_2Cl_2$-CuI, 50 °C; (b) NEt_3, 1-hexyne, $Pd(PPh_3)_2Cl_2$-CuI, 50 °C.

butyl chain and with the 2′-proton on the 4-phenyl group. Consistent with our experience with Suzuki reactions the 4-alkoxy-5-iodopyrimidine (**17**) was a better substrate for a palladium catalysed alkynylation. Although the products from coupling of (**17**) and 1-hexyne could not be separated completely, detection, by ^{1}H NMR, of the novel alkynylpyrimidine **27** (27%) together with recovered starting material **17** (55%), confirmed the potential of this route.

Quantum-Mechanical Calculations

Choice of modelling methods

The quantum-mechanical studies on bropirimine and analogous structures considered the conformational flexibility of the 6-phenyl group, and the terms involved in the formation of a hydrogen-bonded tautomeric duplex, both in gas and solution phases. These terms may be combined into a free energy cycle (Fig. 2). For this work we chose to use semi-empirical PM3 calculations[25] for gas-phase geometry optimizations and heat of formation calculations, combined with the SM3 continuum solvation model for the prediction of free energies of hydration. The PM3 method has been shown to model hydrogen bonding convincingly,[26,27] and the SMx approach has been found to accurately predict free energies of solvation and partition coefficients.[28–31]

Conformational analysis

One possible route by which variation in the 5-substituent in 2-aminopyrimidin-4-ones might affect biological activity is through its effect on the rotational freedom of the 6-phenyl group. The PM3 calculations indicated that up to the point where steric clashes began, the phenyl group showed almost free rotation. The effect of the variation in the bulk of the 5-substituent on the range of dihedral angles available for an energy penalty <5 kcal mol^{-1} is shown in Table 2. All angles are accessible with 5-H and 5-F but the other halogen groups restrict rotation to angles >20–$40°$ depending on their size. A 5-Me substituent has an effect intermediate between 5-Br and 5-I. As predicted, a 5-Ph group restricts the 6-phenyl substituent to angles

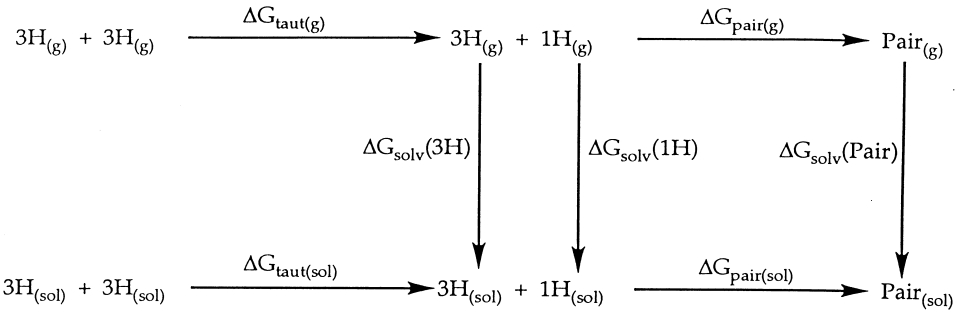
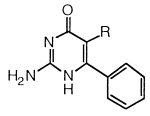
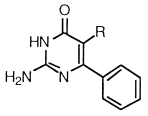


Figure 2. Free energy cycle used to calculate thermodynamic quantities. The quantities are ΔG_{taut}, free energy of tautomerism (3*H* to 1*H*); ΔG_{solv}, free energy of solvation; ΔG_{pair}, free energy of hydrogen-bonded pair formation.

Table 2. Conformational analysis of 2-amino-6-phenyl-5-(substituted)-pyrimidin-4-ones



1*H*-tautomer



3*H*-tautomer

	Angle (°)[a]	
	1*H*-tautomer	3*H*-tautomer
H	0	0
F	0	0
Cl	27	21
Br (bropirimine)	34 (47)[b]	32 (52)[b]
I	39	34
Me	39	30
Ph	70	65

[a]Minimum dihedral angle between the 6-phenyl group and the pyrimidine rings accessible for an energy penalty < 5 kcal mol^{-1}.
[b]Bond angles in brackets are taken from the X-ray crystal structure of the *N*-methylformamide solvate of bropirimine **5** (ref 6).

> 65–$70°$. In all cases the molecular modelling predicts that the 1*H*-tautomers show more restricted rotation (larger dihedral angle) than their 3*H*-counterparts presumably because of the steric clash between the pyrimidine 1*H* and the *ortho* hydrogen on the 6-phenyl residue. This result contrasts with the dihedral angle in the crystallographic structure of bropirimine **5** which is greater in the 3*H*-tautomer (Fig. 1). Possibly perturbations induced by the solvating *N*-methylformamide within the crystal lattice account for these differences.

Thermodynamic analysis of tautomerism

The free energy components of the thermodynamic cycle (Fig. 2) are shown in Table 3. In the gas phase, the 3*H*-tautomers are always predicted to be more stable than the 1*H*-tautomers. This is also observed in solution, but to a lesser extent, as the 1*H*-forms show more favourable free energies of solvation. Amongst the 5-halo derivatives, the relative stability of the 1*H*-tautomers decreases in the order: F > Cl > I > Br. Despite this ranking, even for a 5-Br substituent, it is clear from the crystal structure data on bropirimine[6] that the 1*H*-tautomer remains accessible providing the environmental conditions are suitable. Theoretical predictions of tautomer stabilities of a series of 5,6-diphenyl-pyrimidinones bearing substituents in the *para* position of the 5-phenyl group (Table 3) show the relative stabilities of the 1*H*-tautomers decrease in the order: OMe > NO_2 > H > Cl, with no obvious relationship to the electronic characteristic of the substituent.

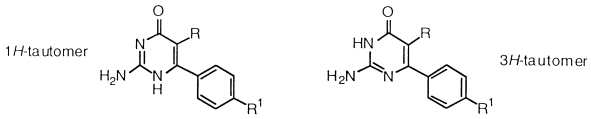
Thermodynamic analysis of base-pairing

On the basis of crystal structure data of bropirimine, the 1*H*-tautomers of the 2-amino-5-(substituted)-6-phenyl-pyrimidin-4-ones can be regarded as being prototypical targets for 'base-pairing' recognition by 3*H*-tautomers, and vice-versa. The calculation of free energies of pairing gives an insight into the strength of this interaction, and how it is affected by changes in the 5- and 6-substituents. In the gas phase, the free energy of pairing for the 5-halo derivatives varies from 0.3 kcal mol^{-1} (for F) to -1.4 kcal mol^{-1} (Cl). For the 5-aryl compounds, both strongly electron-donating (OMe) and withdrawing (NO_2) attached groups disfavour pair formation. In solution, all free energies of pairing are positive because the free energy of solvation of the individual partners is greater than that of the H-bonded pair: the weakest interactions are made by the 5-fluoro and 5-(4-methoxyphenyl) derivatives, and the strongest by the 5-chloro compound.

Conclusions

Attempts to rationalize the broad chemotherapeutic properties of bropirimine and other bio-active molecules bearing the 2-aminopyrimidin-4-one pharmacophore, in terms of their abilities to participate in base-pairing encounters, is thwarted by the fragmented nature of the published bio-data.[2–4] However, the following trends can be discerned: for antitumour activity, bromine is preferred to iodine at C(5), and phenyl to methyl at C(6); for antiviral properties in the 6-phenyl derivatives, activity of the 5-halo series follows the order Cl > Br > I > F and groups smaller than chloro or larger than propyl are disfavoured; when interferon induction is considered as the biological end-point, most data fit the trends observed for antiviral activity.

Table 3. Free energy components in formation of hydrogen-bonded tautomeric pairs of 2-amino-6-phenyl-5-(substituted)pyrimidin-4-ones[a,b]



R	R^1	$G_{f,(g)}$	$\Delta G_{pair,(g)}$	ΔG_{solv}	$\Delta G_{pair,(sol)}$	$\Delta G_{taut,(g)}$	$\Delta G_{taut(sol)}$
H	H	—	−0.8	—	12.2	6.59	3.0
		−7.13(1*H*)		−23.21(1*H*)			
		−13.72(3*H*)		−19.60(3*H*)			
		−19.80(pair)		−29.83(pair)			
F	H		0.3		14.2	4.51	2.6
		−48.74(1*H*)		−24.25(1*H*)			
		−53.22(3*H*)		−22.34(3*H*)			
		−99.81(pair)		−32.69(pair)			
Cl	H		−1.4		12.0	5.48	3.3
		−12.83(1*H*)		−23.03(1*H*)			
		−18.31(3*H*)		−20.90(3*H*)			
		−30.67(pair)		−30.51(pair)			
Br	H		−1.1		12.4	6.26	4.7
		2.61(1*H*)		−23.34(1*H*)			
		−3.65(3*H*)		−21.83(3*H*)			
		−0.28(pair)		−31.71(pair)			
I	H		−0.5		12.1	5.99	4.2
		23.14(1*H*)		−21.85(1*H*)			
		17.15(3*H*)		−20.02(3*H*)			
		41.62(pair)		−29.31(pair)			
Me	H		−0.8		12.1	5.04	2.4
		−17.53(1*H*)		−21.26(1*H*)			
		−22.57(3*H*)		−18.63(3*H*)			
		−39.11(pair)		−26.98(pair)			
Ph	H		−0.3		13.5	6.20	3.7
		15.96(1*H*)		−23.29(1*H*)			
		9.76(3*H*)		−20.82(3*H*)			
		27.21(pair)		−30.34(pair)			
Ph	OMe		0.2		14.2	3.88	1.5
		−26.59(1*H*)		−24.57(1*H*)			
		−30.47(3*H*)		−22.18(3*H*)			
		−55.01(pair)		−32.80(pair)			
Ph	NO_2		−0.1		13.6	5.62	3.1
		2.73(1*H*)		−26.33(1*H*)			
		−2.89(3*H*)		−23.82(3*H*)			
		1.52(pair)		−36.39(pair)			
Ph	Cl		−1.0		13.2	8.40	5.6
		9.93(1*H*)		−24.37(1*H*)			
		1.53(3*H*)		−21.92(3*H*)			
		11.94(pair)		−31.99(pair)			

[a]Values in kcal mol^{-1}.

[b]$\Delta G_{pair,(g)} = G_{f,(g)}(pair) - G_{f,(g)}(1H) - G_{f,(g)}(3H)$; $\Delta G_{pair,(sol)} = \Delta G_{pair,(g)} + \Delta G_{solv}(pair) - \Delta G_{solv}(3H) - \Delta G_{solv}(1H)$; $\Delta G_{taut,(g)} = G_{f,(g)}(1H) - G_{f(g)}(3H)$; $\Delta G_{taut(sol)} = \Delta G_{taut,(g)} + \Delta G_{f,(sol)}(1H) - \Delta G_{f,(sol)}(3H)$.

In the present study we have considered a range of compounds with differing conformational and electronic characteristics which may contribute to their recognition capabilities, possibly at specific DNA sequences. The most bio-active agents identified in the literature are bropirimine **5** and its 5-chloro congener which are also calculated to have a propensity for strong H-bonding interactions. An ability to adopt the less favoured 1*H*-tautomeric arrangement does not seem to be a critical factor. However, the structural characteristics optimizing H-bonding in 2-amino-5-halo-6-phenylpyrimidin-4-ones in general also dictate that this type of molecule is almost totally insoluble in water since the base-pairing phenomenon in the solid state effectively presents only hydrophobic halogen and phenyl groups to the aqueous environment.[11] Overall, despite its unfavourable pharmaceutical characteristics, the modelling data supports the selection of bropirimine **5** as the agent of clinical choice.

In our synthetic work we have shown that sterically-hindered 2-amino-5-halo-6-phenylpyrimidin-4-ones are only poor substrates for palladium catalysed couplings but that 2-amino-5-iodo-4-methoxy-6-phenylpyrimidine (**17**) can be converted efficiently to 5-aryl derivatives employing, optimally, a combination of an arylboronic acid, $Pd(dppf)(OAc)_2$ catalyst and tripotassium phosphate base in aqueous 1,2-dimethoxyethane as solvent. The 4-methoxypyrimidines can then be processed hydrolytically to the required 2-amino-5-aryl-6-phenylpyrimidin-4-ones. This process offers a route to novel structures which may facilitate a more detailed SAR analysis of this intriguing series of compounds.

Experimental

General methods

All melting points were recorded on a Gallenkamp melting point apparatus, and are uncorrected. IR spectra were taken on a Mattson 2020 GALAXY Series FT-IR spectrometer as KBr discs. ^{1}H, ^{13}C spectra were recorded on a Bruker ARX250 spectrometer at 250.1 and 62.9 MHz, respectively, in solvents as specified, with tetramethylsilane or residual protic solvents as internal standard, *J* values being in Hz. Low resolution mass spectra were recorded on a Micromass Platform (AP^+, ES^+). High resolution mass spectrometry (HRMS) were performed by the EPSRC Mass Spectrometry Service Centre, Swansea, UK. Silica gel TLC was performed on 60F-254 pre-coated sheets (E. Merck) and column chromatography on silica gel C60 (60–120 mesh). Elemental analyses were performed by Knoll Pharmaceuticals Physical Chemistry Department, Nottingham, UK. All solvents used in palladium catalysed cross-coupling reactions were purged with nitrogen before use.

Arylation of 2-amino-5-halo-6-(substituted)pyrimidin-4-ones by cross-coupling reactions. Results of cross-coupling reactions between 2-amino-5-bromo-6-phenylpyrimidin-4(3*H*)-one (**5**) and 2-amino-5-iodo-6-phenylpyrimidin-4(3*H*)-one **6** and benzeneboronic acid under a range of conditions are summarised in Table 1 (Entries 1–9). Products were isolated by silica gel column chromatography, with ethyl acetate:methanol:acetic acid (16:4:1) as eluent, or the mixed pyrimidinone products were analysed by ^{1}H NMR spectroscopy in [$^2H_6$]DMSO. Recorded below are details of syntheses which gave fully characterised products.

2-Amino-6-methyl-5-phenylpyrimidin-4(3*H*)-one (11). 2-Amino-5-bromo-6-methylpyrimidin-4(3*H*)-one (**7**) (0.414 g, 2.03 mmol) and tetrakis(triphenylphosphine)-palladium(0) (0.080 g, 0.07 mmol) were stirred in DMF (5 mL) for 5 min. Benzeneboronic acid (0.275 g, 2.25 mmol) and more DMF (10 mL) were added, followed by sodium carbonate (0.550 g, 5.19 mmol) dissolved in water (2 mL) and the mixture was heated to reflux for 24 h, under a gentle flow of nitrogen. The cooled, filtered, mixture was evaporated in vacuo. The residue was purified by silica gel column chromatography, with ethyl acetate:methanol:acetic acid (16:4:1) as eluent to yield 2-amino-6-methylpyrimidin-4(3*H*)-one (**3**) (85%) and the pyrimidinone **11** (0.040 g, 10%) as a white solid with R_f 0.61. Compound **11** crystallised from ethanol with mp >250 °C (decomp.); ν_{max} (KBr)/cm^{-1} 3368, 3134, 2925, 1655, 1613, 1508, 1048 and 704; δ_H ([$^2H_6$]DMSO) 1.98 (3H, s, Me), 6.96 (2H, br s, NH_2), 7.18–7.40 (5H, m, Ph) and 10.5–12.5 (1H, br s, NH); δ_C ([$^2H_6$]DMSO) 20.8 (Me), 114.1 (C-5), 126.8 (CH, ArC), 127.8 (2×CH, ArC), 130.6 (2×CH, ArC), 134.4 (C, ArC), 153.1 (C), 157.0 (C) and 161.9 (C); HRMS calcd for $C_{11}H_{11}N_3O$ 201.0902, found 201.0899. Anal. calcd for $C_{11}H_{11}N_3O$: C, 65.6; H, 5.7; N, 20.7. Found: C, 65.7; H, 5.5; N, 20.9%.

Using the conditions of Wang and Haseltine,[14] which employ toluene–ethanol as a solvent and aqueous sodium carbonate (2 mol equiv) as a base, only a trace of coupling (TLC) was observed and >90% starting material **7** was recovered.

2-Amino-5,6-diphenylpyrimidin-4(3*H*)-one (12). Using the conditions described (above) for the synthesis of pyrimidinone **11**, the bromopyrimidinone **5** was converted to the debrominated pyrimidinone **2** (83%) and the required diphenylpyrimidinone **12** (17%). A pure sample of **12** had mp 312–314 °C (lit.,[7] mp 319 °C); δ_H ([$^2H_6$]DMSO) 6.64 (2H, br s, NH_2), 6.86–7.02 (2H, m, ArH), 7.06–7.22 (8H, m, ArH) and 11.12 (1H, br s, NH); δ_C([$^2H_6$]DMSO) 113.2 (C-5), 126.1 (CH, ArC), 127.5 (2×CH, ArC), 127.6 (2×CH, ArC), 128.2 (CH, ArC), 129.4 (2×CH, ArC), 131.6 (2×CH, ArC), 135.7 (C, ArC), 139.7 (C, ArC), 154.3 (C), 162.1 (C) and 162.7 (C); HRMS calcd for $C_{16}H_{13}N_3O$ 263.1058, found 263.1038.

Synthesis of 2,4-disubstituted-5-halo-6-phenylpyrimidines. The following pyrimidines were prepared.

2-Amino-5-bromo-4-methoxy-6-phenylpyrimidine (14). Bropirimine **5** (3.725 g, 14.0 mmol) was heated to reflux in phosphorous oxychloride (12 mL) for 45 min. Following removal of the excess oxychloride in vacuo the residue was triturated in aqueous ammonia-ice and the solid collected. The product was warmed in dichloromethane (50 mL) and an insoluble by-product was filtered off. Evaporation of the dried ($MgSO_4$) dichloromethane afforded 2-amino-5-bromo-4-chloro-6-phenylpyrimidine **13** (2.68 g, 67%) as yellow crystals, mp 135 °C (lit.,[12] mp 136–138 °C).

A suspension of **13** (0.65 g) in anhydrous methanol (25 mL) was refluxed (0.5 h) under nitrogen with a 25% solution of sodium methoxide in methanol (2 mL). Excess sodium methoxide was decomposed by solid carbon dioxide and the mixture was diluted with water (200 mL). The suspension was extracted with chloroform (3×50 mL) and the combined organic extracts were washed with water (25 mL), dried ($MgSO_4$) and reduced to give a white solid. The solid was crystallised from hexane to furnish the methoxypyrimidine **14** (0.588 g, 93%), mp 174 °C; ν_{max} (KBr)/cm^{-1} 3486, 3291, 3154, 1630, 1543, 1375, 1200 and 698; δ_H($CDCl_3$) 4.01 (3H, s, Me), 5.11 (2H, br s, NH_2), 7.40–7.47 (3H, m, ArH) and 7.61–7.65 (2H, m, ArH); δ_C($CDCl_3$) 54.8 (Me), 92.5 (C-5), 127.9 (2×CH, ArC), 128.8 (2×CH, ArC), 129.2 (CH, ArC), 138.1 (C, ArC), 160.9 (C), 166.0 (C) and 166.8 (C); *m*/*z* M^+ (ES^+), 280, 282. Anal. calcd for $C_{11}H_{10}BrN_3O$: C, 47.2; H, 3.6; N, 15.0. Found: C, 47.1; H, 3.5; N, 14.8%.

2-Amino-5-iodo-4-methoxy-6-phenylpyrimidine (17). 2-Amino-4-methoxy-6-phenylpyrimidine **16** (1.29 g, 6.41 mmol), prepared from 3,3-bis(methylthio)-1-phenyl-2-propene-1-one and guanidine sulphate,[17] was heated to reflux with *N*-iodosuccinimide (1.88 g, 8.33 mmol) in anhydrous chloroform (20 mL) for 2 h, under nitrogen. After cooling, the red solution was diluted with more chloroform (30 mL) and washed with water (2×25 mL), 5% sodium metabisulfite solution (15 mL), water (10

mL), then dried ($MgSO_4$) and concentrated to dryness in vacuo to provide the methoxypyrimidine **17** (2.07 g, 99%) as a pale yellow solid, mp 179–180 °C (from hexane); ν_{max} (KBr)/cm^{-1} 3476, 3150, 1628, 1545, 1362, 1198, 1005 and 696; δ_H($CDCl_3$) 3.96 (3H, s, Me), 5.46 (2H, br s, NH_2) and 7.42–7.54 (5H, m, Ph); δ_C ($CDCl_3$) 54.9 (Me), 64.9 (C-5), 127.9 (2×CH, ArC), 128.6 (2×CH, ArC), 129.0 (CH, ArC), 140.5 (C, ArC), 162.2 (C), 168.7 (C) and 170.2 (C); *m/z* M^+(AP^+), 328. Anal. calcd for $C_{11}H_{10}IN_3O$: C, 40.3; H, 3.0; N, 12.9. Found: C, 40.7; H, 3.0; N, 12.9%.

Arylation of 2-amino-5-halo-4-methoxy-6-phenylpyrimidines by cross-coupling reactions. A summary of the results of cross-coupling reactions between 2-amino-5-bromo-4-methoxy-6-phenylpyrimidin-4(3*H*)-one (**14**) and 2-amino-5-iodo-4-methoxy-6-phenylpyrimidin-4(3*H*)-one (**17**) and benzeneboronic acids under a range of conditions are recorded in Table 1 (Entries 10–21). Listed below are details of syntheses which gave the optimum yields of fully characterised products.

2-Amino-4-methoxy-5,6-diphenylpyrimidine (18). A solution of sodium carbonate (0.276 g, 2.61 mmol) in water (2 mL) was added to a mixture of 2-amino-5-bromo-4-methoxy-6-phenylpyrimidine **14** (0.287 g, 1.02 mmol), benzeneboronic acid (0.138 g, 1.13 mmol) and tetrakis(triphenylphosphine)palladium(0) (0.040 g, 0.035 mmol) in toluene (2 mL) and ethanol (2 mL) and the mixture was heated to reflux for 18 h, under nitrogen. After cooling, the mixture was extracted with chloroform (3×25 mL), and the combined extracts were washed successively with water (10 mL), 0.5 N-sodium hydroxide (2×5 mL), water (10 mL) and saturated brine (10 mL). The organic layer was then dried ($MgSO_4$) and evaporated to give a grey solid which was crystallised from 10% ethyl acetate–hexane to yield the pyrimidine **18** (0.212 g, 75%), mp 185–185.5 °C as a crystalline white solid; ν_{max}(KBr)/cm^{-1} 3379, 3171, 1638, 1568, 1543, 1371, 1198 and 696; δ_H($CDCl_3$) 3.88 (3H, s, Me), 5.14 (2H, br s, NH_2) and 7.08–7.28 (10H, m, 2×Ph); δ_C($CDCl_3$) 53.9 (Me), 110.9 (C-5), 126.7 (CH, ArC), 127.7 (2×CH, ArC), 127.8 (2×CH, ArC), 128.3 (CH, ArC), 129.4 (2×CH, ArC), 131.2 (2×CH, ArC), 134.4 (C, ArC), 138.6(C, ArC), 161.3 (C), 165.2 (C) and 168.5 (C); *m/z* M^+(ES^+), 278. Anal. calcd for $C_{17}H_{15}N_3O$: C, 73.6; H, 5.4; N, 15.0. Found: C, 73.5; H, 5.4; N, 15.0%.

2-Amino-5-(2,4-dichlorophenyl)-4-methoxy-6-phenylpyrimidine (19). Prepared as above, from **14** (0.210 g, 0.75 mmol) and 2,4-dichlorobenzeneboronic acid (0.158 g, 0.83 mmol), tetrakis(triphenylphosphine)palladium(0) (0.030 g, 0.026 mmol) and sodium carbonate (0.203 g, 1.92 mmol) in refluxing toluene–aqueous ethanol (24 h), the crude reaction mixture was subjected to three sequential silica gel column purifications with chloroform–diethyl ether (85:15) as eluent and gave a white solid (0.093 g) that by ^{1}H NMR was a 1:1 mixture of **19** and starting pyrimidine **14**. Crystallisation of this mixture from acetone (1 mL) gave a sample 90% enriched in **19** (0.021 g); ν_{max}(KBr)/cm^{-1} 3408, 3183, 1645, 1568, 1368, 1051, 814 and 698; δ_H($CDCl_3$) 3.89 (3H, s, Me), 5.23 (2H, br s, NH_2), 6.92 (1H, d, J=8, 6′-H (5-Ar)), 7.09 (1H, dd, J=8 and 2, 5′-H (5-Ar)), 7.17–7.31 (5 H, m, Ph) and 7.40 (1H, d, J=2, 3′-H (5-Ar)); δ_C($CDCl_3$) 54.0 (Me), 107.5 (C-5), 126.9 (CH, ArC), 127.9 (2×CH, ArC), 128.6 (2×CH, ArC), 128.7 (CH, ArC), 129.1 (CH, ArC), 132.8 (C, ArC), 133.6 (CH, ArC), 133.7 (C, ArC), 136.1(C, ArC), 138.1 (C, ArC), 162.1 (C), 165.7 (C) and 168.4 (C); MS (ES^+) calcd for $C_{17}H_{13}Cl_2N_3O$ 346, 348, 350, found 346, 348, 350.

2-Amino-4-methoxy-6-phenyl-5-(3-thienyl)pyrimidine (20). Palladium acetate (5.1 mg, 0.023 mmol) and 1,1′-(diphenylphosphino)ferrocene (dppf) (16.6 mg, 0.03 mmol) were heated in degassed 1,2-dimethoxyethane (DME) (3 mL) for 15 min, under nitrogen. To the cooled mixture 2-amino-5-bromo-4-methoxy-6-phenylpyrimidine (**14**) (0.210 g, 0.75 mmol), thiophene-3-boronic acid (0.106 g, 0.83 mmol) and tripotassium phosphate (0.318 g, 1.5 mmol) were added, along with DME (3 mL) and water (1 mL), and the mixture was heated under gentle reflux for 22 h, under nitrogen. The reaction mixture was cooled, diluted with water (10 mL) and extracted with diethyl ether (3×25 mL). The combined ethereal extracts were washed with water (10 mL), saturated brine (10 mL), dried ($MgSO_4$) and reduced in vacuo to leave a pale brown solid. Succesive crystallisations of the brown solid from hexane–ethyl acetate (5:1), then acetone, yielded the thienylpyrimidine **20** (0.055 g, 26%) as a crystalline white solid, mp 206–207 °C; ν_{max}(KBr)/cm^{-1} 3461, 3144, 1630, 1549, 1479, 1452, 1344 and 1198; δ_H($CDCl_3$) 3.93 (3H, s, Me), 4.99 (2H, br s, NH_2), 6.82 (1H, dd, J=1 and 5, 4′-H (thienyl)), 6.96 (1 H, dd, J=1 and 3, 2′-H (thienyl)), 7.18 (1H, dd, J=3 and 5, 5′-H (thienyl)) and 7.23–7.32 (5 H, m, Ph); δ_C($CDCl_3$) 53.9 (Me), 115.4 (C-5), 124.7 (CH, thienyl), 125.2 (CH, thienyl), 128.2 (2×CH, ArC), 129.0 (CH), 130.2 (2×CH, ArC), 131.2 (CH), 135.3 (C), 140.4 (C), 162.9 (C), 166.2 (C) and 169.3 (C). Anal. calcd for $C_{15}H_{13}N_3OS$: C, 63.6; H, 4.55; N, 14.8. Found: C, 63.6; H, 4.55; N, 14.6%.

2-Amino-4-methoxy-5-(4-methoxyphenyl)-6-phenylpyrimidine (21). Prepared according to the method for the preparation of **20**, with palladium acetate (5.1 mg, 0.023 mmol), dppf (16.6 mg, 0.03 mmol), 2-amino-5-iodo-4-methoxy-6-phenylpyrimidine (**17**) (0.245 g, 0.75 mmol), 4-methoxybenzeneboronic acid (0.126 g, 0.83 mmol) and tripotassium phosphate (0.318 g, 1.5 mmol), in DME (6 mL) and water (1 mL), with a reaction time of 24 h. The crude residue from the organic extract was purified by silica gel chromatography, with chloroform: diethyl ether (4:1) as eluent, to provide impure **21** (0.20 g). Crystallisation from acetone yielded white crystals (0.165 g, 72%), mp 174 °C; ν_{max}(KBr)/cm^{-1} 3484, 3140, 1545, 1371, 1246, 1042, 831 and 696; δ_H($CDCl_3$) 3.78 (3H, s, Me), 3.90 (3H, s, Me), 5.05 (2H, br s, NH_2), 6.76–6.80 (2H, m, ArH), 7.00–7.04 (2H, m, ArH) and 7.19–7.31 (5H, m, Ph); δ_C($CDCl_3$) 54.0 (Me), 55.1 (Me), 110.5 (C-5), 113.4 (2×CH, ArC), 126.4 (C, ArC), 127.8 (2×CH, ArC), 128.2 (CH, ArC), 129.4 (2×CH, ArC), 132.2 (2×CH, ArC), 138.8 (C, ArC), 158.3 (C), 161.1 (C), 165.1 (C) and 168.7 (C). Anal. calcd for $C_{18}H_{17}N_3O_2$: C, 70.3; H, 5.6; N, 13.7. Found: C, 70.4; H, 5.5; N, 13.55%.

2-Amino-5-(4-chlorophenyl)-4-methoxy-6-phenylpyrimidine (22). Prepared according to the method for the preparation of **20**, with palladium acetate (5.1 mg, 0.023 mmol), dppf (16.6 mg, 0.03 mmol), 2-amino-5-iodo-4-methoxy-6-phenylpyrimidine (**17**) (0.245 g, 0.75 mmol), 4-chlorobenzeneboronic acid (0.130 g, 0.83 mmol) and tripotassium phosphate (0.318 g, 1.5 mmol), in DME (6 mL) and water (1 mL), with a reaction time of 16 h. The crude reaction product was crystallised from hexane:acetone (approx. 2:1) to afford **22** (0.182 g, 78%) as pale brown crystals, mp 196–198 °C; ν_{max}(KBr)/cm^{-1} 3493, 3140, 1634, 1566, 1543, 1370, 833 and 694; δ_H($CDCl_3$) 3.90 (3H, s, Me), 5.16 (2H, br s, NH_2), 7.01–7.06 (2H, m, 2×ArH) and 7.17–7.30 (7H, m, ArH); δ_C($CDCl_3$) 53.7 (Me), 107.5 (C-5), 127.7 (2×CH, ArC), 127.9 (2×CH, ArC), 128.4 (CH, ArC), 129.4 (2×CH, ArC), 131.2 (C, ArC), 133.1 (2×CH, ArC), 134.0 (C, ArC), 138.8 (C, ArC), 162.2 (C), 164.9 (C) and 167.8 (C). Anal. calcd for $C_{17}H_{13}ClN_3O$: C, 65.5; H, 4.5; N, 13.5. Found: C, 65.6; H, 4.75; N, 13.4%.

2-Amino-4-methoxy-5-(3-nitrophenyl)-6-phenylpyrimidine (23). Prepared according to the method for the preparation of **20**, with palladium acetate (5.1 mg, 0.023 mmol), dppf (16.6 mg, 0.03 mmol), 2-amino-5-iodo-4-methoxy-6-phenyl-pyrimidine (**17**) (0.245 g, 0.75 mmol), 3-nitrobenzeneboronic acid (0.139 g, 0.83 mmol) and tripotassium phosphate (0.318 g, 1.5 mmol), in DME (6 mL) and water (1 mL), with a reaction time of 16 h. Crystallisation of the crude reaction product from hexane:acetone (approx. 5:2) yielded **23** (0.197 g, 75%) as its 2:1 solvate with acetone, mp 184–185 °C; ν_{max} (KBr)/cm^{-1} 3322, 3198, 1640, 1545, 1346, 1200, 1065 and 775; δ_H($CDCl_3$) 2.18 (3H, s, Me, acetone), 3.91 (3H, s, Me), 5.24 (2H, br s, NH_2), 7.16–7.31 (5H, m, Ph), 7.32–7.39 (2H, m, ArH) and 8.00–8.09 (2H, m, ArH); δ_C($CDCl_3$) 30.9 (Me–acetone), 54.1 (Me), 108.6 (C-5), 121.6 (CH, ArC), 126.2 (CH, ArC), 128.2 (2×CH, ArC), 128.6 (CH, ArC), 128.8 (CH, ArC), 129.4 (2×CH, ArC), 136.5 (C, ArC), 137.5 (CH, ArC), 137.8 (C, ArC), 147.9 (C, ArC), 161.7 (C), 165.9 (C), 168.2 (C) and 207 (CO, acetone). Anal. calcd for $C_{17}H_{13}N_4O_3$: C, 63.2; H, 4.8; N, 15.9. Found: C, 63.0; H, 4.8; N, 16.0%.

Hydrolysis of 2-amino-5,6-diphenyl-4-methoxypyrimidine (18). 10 N-Hydrochloric acid (5 mL) was added to a solution of 2-amino-5,6-diphenyl-4-methoxypyrimidine (**18**) (0.044 g, 0.16 mmol) in methanol (5 mL), and the mixture was heated to reflux for 1 h. After cooling, the solution was basified with solid sodium hydroxide (2.27 g) and then re-acidified with acetic acid to pH 6. The precipitate was collected, washed with water and dried in vacuo to furnish the pyrimidinone **12** (0.040 g, 96%) as a white solid, mp 314–316 °C (from ethanol) identical (^{1}H NMR) to the sample prepared by a palladium catalysed coupling between the bromopyrimidinone **5** and benzeneboronic acid (see earlier).

Hydrolysis of 2-amino-4-methoxy-5-(4-methoxyphenyl)-6-phenylpyrimidine (21). The 6-phenylpyrimidine (**21**) (0.17 g, 0.55 mmol) was heated to reflux in a mixture of methanol (5 mL) and 10 N-hydrochloric acid (5 mL) for 2 h. After cooling the reaction mixture was diluted with water (10 mL), basified with solid sodium hydroxide (2.24 g) and re-acidified with acetic acid to pH 6. The precipitate was collected, washed with water followed by a small portion of acetone, and dried to furnish **24** (0.16 g, 97%) which formed white crystals, mp 322–326 °C (from ethanol–ethyl acetate); ν_{max}(KBr)/cm^{-1} 3383, 3108, 1651, 1582, 1518, 1242, 993 and 704; δ_H([$^2H_6$]DMSO) 3.68 (3H, s, Me), 6.61 (2H, br s, NH_2), 6.68–6.75 (2H, m, ArH), 6.89–6.95 (2H, m, ArH), 7.16–7.23 (5H, m, Ph) and 11.10 (1H, br s, NH); δ_C([$^2H_6$]DMSO) 55.1 (Me), 112.9 (C-5), 113.1 (2×CH, ArC), 127.5 (2×CH, ArC), 127.7 (C, ArC), 128.1 (CH, ArC), 129.4 (2×CH, ArC), 132.5 (2×CH, ArC), 139.9 (C, ArC), 154.1 (C), 157.6 (C), 161.7 (C) and 162.9 (C). Anal. calcd for $C_{17}H_{15}N_3O_2$: C, 69.2; H, 5.2; N, 14.2. Found: C, 60.1; H, 5.4; N, 14.1%.

Alkynylations of pyrimidines (6) and (17). 2-Amino-6-butyl-4-phenylfuro[2,3-*d*]pyrimidine (26). Triethylamine (8 mL) was added to a stirred suspension of 2-amino-5-iodo-6-phenylpyrimidin-4(3*H*)-one (**6**) (0.626 g, 2.00 mmol), bis(triphenylphosphine)palladium dichloride (0.028 g, 0.04 mmol) and cuprous iodide (0.100 g, 0.52 mmol) in DMF (8 mL) under nitrogen. A deep green solution was formed. 1-Hexyne (0.329 g, 4.00 mmol) was added and the mixture was stirred at room temperature for 40 h. The reaction mixture was then heated to 50 °C for 4 h. Concentration of the reaction mixture in vacuo gave a brown solid which was suspended in diethyl ether (50 mL) and the organic layer was washed successively with 1 N-sodium hydroxide (2×10 mL), water (10 mL), 1 N-hydrochloric acid (2×10 mL), water (10 mL) and saturated brine (10 mL). The ethereal layer was dried ($MgSO_4$) and reduced in vacuo to give a brown oil, which was purified by silica gel column chromatography with diethyl ether:dichloromethane (1:10), to give a sample of the fluorescent furopyridine (**26**) (0.021 g, 4%): ν_{max} ($CDCl_3$ film)/cm^{-1} 3530, 3422, 2961, 2934, 1616, 1460, 1371 and 698; δ_H($CDCl_3$) 0.95 (3H, t, J = 7, $CH_2CH_2CH_2CH_3$), 1.43 (2H, quintet, J = 7, $CH_2CH_2CH_2CH_3$), 1.68–1.79 (2H, m, $CH_2CH_2CH_2CH_3$), 2.74 (2H, td, J = 7 and 1, $CH_2CH_2CH_2CH_3$), 5.20 (2H, br s, NH_2), 6.52 (1H, t, J = 1, H-5), 7.47–7.56 (3H, m, ArH) and 7.95–8.01 (2H, m, ArH).

2-Amino-5-(1-hexyn-1-yl)-4-methoxy-6-phenylpyrimidine (27). 1-Hexyne (0.14 mL, 1.2 mmol) was added to a mixture of 2-amino-5-iodo-4-methoxy-6-phenylpyrimidine (**17**) (0.327 g, 1.0 mmol), bis(triphenylphosphine)palladium dichloride (0.014 g, 0.02 mmol) and cuprous iodide (0.050 g, 0.26 mmol) in degassed (argon) triethylamine (10 mL). The reaction mixture was then heated to 50–60 °C for 16 h, concentrated to dryness in vacuo, suspended in water (50 mL) and the mixture extracted with dichloromethane (3×25 mL). The combined organic extracts were washed with water (10 mL), dried ($MgSO_4$) and evaporated to leave an oily brown solid. Silica gel column purification of the crude product with diethyl ether–chloroform (1:10) as eluent, gave a band of R_f 0.36 (0.255 g, 82% recovery) comprising starting pyrimidine **17** and the alkynylpyrimidine **27** (2:1). The ^{1}H NMR spectrum of the weakly fluorescent alkyne **27** (at 254 nm) showed δ_H($CDCl_3$) 1.22 (3H, t,

$J=7$, $CH_2CH_2CH_2CH_3$), 1.26–1.56 (4H, m, $CH_2CH_2CH_2CH_3$), 2.38 (2H, t, $CH_2CH_2CH_2CH_3$), 3.97 (3H, s, OMe), 5.57 (2H, br s, NH_2), 7.39–7.45 (3H, m, ArH) and 7.88–7.92 (2H, m, ArH).

Molecular modelling studies

The structure of bropirimine **5** was taken from the reported X-ray crystal structure.[6] The (solvating) *N*-methylformamide in the unit cell was removed and initial geometry optimizations were started on the pyrimidinone base-pair. Individual tautomer structures were also extracted from this structure. Analogues of bropirimine were constructed by substituting appropriate atoms or functional groups at the 5- and 6-positions. All structures were geometry optimized in the gas phase at the PM3 level. and calculations run on Silicon Graphics workstations using SPARTAN 4.0.[32]

Gas heats of formation (HOF) were corrected for vibrational entropy and enthalpy, and the zero point energy (ZPE) was subtracted to arrive at a value for $\Delta G_{(g)}$ (eq. (1)). The vibrational values were calculated at 298.15 K from a frequency analysis within SPARTAN. After a solvation calculation was run using SM3, the reported energy (E_{SM3}) was a combination of the gas heat of formation plus the solvation free energy. Subtraction of the heat of formation from the SM3 energy provides ΔG_{solv} (eq. (2)). Solution phase ΔG values (ΔG_{sol}) were obtained by adding $\Delta G_{(g)}$ and ΔG_{solv} and correcting for the differences in standard states (eqs (3) and (4)).[33]

$$\Delta G_{(g)} = \mathrm{HOF} + H_{vib} + H_{rot} + H_{tran} - ZPE - [T(S_{vib} + S_{rot} + S_{tran})] \quad (1)$$

$$\Delta G_{solv} = E_{SM3} - \Delta G_{(g)} \quad (2)$$

$$\alpha = \mathrm{RTln}(1/22.4\mathrm{L}) = -1.83\,\mathrm{kcal\,mol^{-1}} \quad (3)$$

$$\Delta G_{(sol)} = \Delta G_{(g)} + \Delta G_{solv} + \alpha \quad (4)$$

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